

# **Acta Agronomica Hungarica**

**VOLUME 38, NUMBERS 1-2, 1989**

**EDITOR-IN-CHIEF**

**I. TAMÁSSY**

**EDITOR**

**Á. MÁTHÉ**

**EDITORIAL BOARD**

**S. RAJKI (Vice chairman), I. DIMÉNY, B. GYÖRFFY, A. HORN,  
Z. KIRÁLY, P. KOZMA, E. KURNIK, I. LÁNG, I. MÁTHÉ,  
I. SZABOLCS**



**Akadémiai Kiadó, Budapest**

ACTA AGRONOMICA HUNG. HU ISSN 0238-0161



# ACTA AGRONOMICA

## A QUARTERLY OF THE HUNGARIAN ACADEMY OF SCIENCES

---

*Acta Agronomica* publishes papers in English on agronomical subjects, mostly on basic research.

*Acta Agronomica* is published in yearly volumes of four issues by

AKADÉMIAI KIADÓ

Publishing House of the Hungarian Academy of Sciences

H-1054 Budapest, Alkotmány u. 21.

Manuscripts and editorial correspondence should be addressed to

*Acta Agronomica*

H-1118 Budapest, P.O. Box 53

*Subscription information*

Orders should be addressed to

KULTURA Foreign Trading Company

H-1389 Budapest P.O. Box 149

or to its representatives abroad

---

*Acta Agronomica Hungarica* is abstracted/indexed in AGRICOLA, Biological Abstracts, Bibliography of Agriculture, Chemical Abstracts, Current Contents-Agriculture, Biology and Environmental Sciences, Excerpta Medica, Horticultural Abstracts, Hydro-Index, Plant Breeding Abstracts, Nutrition Abstracts and Reviews

---

© Akadémiai Kiadó, Budapest

## CONTENTS

### SOIL SCIENCE AND AGROCHEMISTRY

Application of electro-ultrafiltration (EUF) method in soil phosphorus determination <i>M. Iqbal Makhdum, M. Nawaz, A. Malik and Fazal Illahi</i> .....	3
Distribution of native fixed ammonium and its prediction as based on textural components and organic carbon in some subtropical soils of West Bengal and Bihar (India) <i>A. Das and B. Datta</i> .....	9

### PLANT PHYSIOLOGY AND BIOCHEMISTRY

Effect of magnesium ion on the anion uptake of plants <i>S. A. Kiss</i> .....	23
Study of flowering in pepper ( <i>Capsicum annuum</i> L.) grown under controlled environments (phytotron) <i>A. Máthé and K. L. Bahadli</i> .....	31
Ways of predicting storage losses in apples <i>P. Sass and Z. Lakner</i> .....	37
Investigation of the chemical composition-changes in horticultural plants as a function of X-ray stimulation doses <i>A. S. Szabó and M. A. J. Tejeda</i> .....	45
Effect of ionizing radiation on the respiration intensity of pears during storage <i>Mahfouz Al Bachir and P. Sass</i> .....	49
Tolerance of five oil crops to salinity and temperature stresses during germination <i>F. S. El Nakhlay and M. A. El Fawal</i> .....	59
Effect of kinetin and salinity on osmotic pressure and carbohydrate contents in two crop plants <i>F. M. Salama and A. A. Awadalla</i> .....	67
Interactive effects of water stress, temperature and $\text{NO}_3^-$ concentration on allocation of soluble nitrogen in germinating <i>Bauhinia</i> seeds <i>H. M. El-Sharkawi and K. A. Farghali</i> .....	77

### PLANT CULTIVATION

Phytomass production of medicinal plants in Finland <i>B. Galambosi</i> .....	89
Effects of plot area and <i>Azolla inocula</i> on growth and nitrogen yield of <i>Azolla pinnata</i> when intercropped with rice <i>D. P. Singh and P. K. Singh</i> .....	99

### PLANT GENETICS AND BREEDING

Heterosis and path coefficient analysis in sesame ( <i>Sesamum indicum</i> L.) <i>H. E. Osman</i> .....	105
--	-----



Utilization of chlorophyll and leaf mutans in $F_1$ hybrids of watermelons and muskmelons <i>K. Mozsár, H. T. Minch and N. Tamássy</i> .....	113
The influence of repeated back-crosses on the productivity of cytoplasmic male sterile lucerne genotypes <i>B. Nagy</i> .....	119
Inheritance of seed colour in mustard <i>D. S. Rawat</i> .....	127
Studies on the agronomic and breeding potentials of some interspecific hybrids in <i>Arachis</i> species <i>K. O. Marfo</i> .....	131

## PLANT PROTECTION

Incidence of <i>Tarsonemina</i> species in soil samples of certain tomato varieties <i>S. M. Abo-Korah and A. A. Younes</i> .....	139
Tomato transplanting and soil <i>Tarsonemina</i> species <i>S. M. Abo-Korah and A. A. Younes</i> .....	143

## ANIMAL PRODUCTION AND GENETICS

Studies on protein utilization of alkaloid-free <i>Luteus albus</i> seed <i>J. Jécsai, M. Szelényi-Galántai and B. Juhász</i> .....	149
--	-----

## LECTURES

Recent results in melon breeding <i>K. Mozsár</i> .....	159
Influence of etological factors in establishing the genetic value of mechanical milkability <i>L. Szajkó</i> .....	163

## REVIEW

Developments in the trace element research <i>I. Pais</i> .....	167
--	-----

BOOK REVIEWS	177
--------------	-----

## *Soil science and agrochemistry*

### APPLICATION OF ELECTRO-ULTRAFILTRATION (EUF) METHOD IN SOIL PHOSPHORUS DETERMINATION

M. IQBAL MAKDHUM, M. NAWAZ, A. MALIK and FAZAL ILLAHI CHOUDHRY

CENTRAL COTTON RESEARCH INSTITUTE, MULTAN, PAKISTAN

(Received: 22 June 1987; accepted 26 August 1987)

The EUF method has established itself as a useful tool for the evaluation of soil fertility. It differs from most other methods of soil analysis in that it not only yields extractable quantities of nutrients but also the rates with which these quantities are desorbed when an external force is applied. The EUF-P values can best be closely correlated with conventional  $\text{NaHCO}_3$  extracted phosphorus (Olsen's method), provided dilution factor is accounted for in calculation of EUF-P in Nemeth's formula.

**Keywords:** Electro-ultrafiltration, EUF-P fractions, rapid determination, soil analysis

#### Introduction

The possibility of determining the amount of effectively available and exchangeable nutrients and the desorbing rate of exchangeable nutrients in the soil by means of Electro-ultrafiltration (EUF) method of Nemeth (1972) has been noted with great interest by many soil scientists. Knowledge of such characteristics as EUF-P, K, Na and Ca nutrients dynamics (Nemeth and Makhdum 1981) provides a possibility for considering the dynamics nature of those plant nutrients that are essential for improved regulation of the nutrient supply to the cultivated crops.

Following the acquisition of an EUF equipment by the Plant Physiology and Chemistry Section, plans were made to test the EUF method's applicability in the soil analysis and fertilizer recommendations. As a first step, the precision of the method was investigated. EUF extraction removes only certain fractions of the total nutrient content in the soil. These fractions are not absolute quantities in the same way as the total content; the amount of the fraction released depends on the voltage applied and desorption time. Therefore, studies on the standardization of the EUF method and its comparison with conventional method were carried out to find its suitability in routine soil phosphorus analysis as a test element.



## Materials and methods

The river sand was collected and treated with concentrated hydrochloric and nitric acid and washed with distilled water to remove all possible nutrients.

Five grammes washed sand sole sample and 5 g sand plus 5 ml of 5 ppm-P of  $\text{KH}_2\text{PO}_4$  was added in the EUF-extraction chamber and was analysed (Pauuw van der, 1969) for P concentration.

As for applicability of EUF to soils, the samples were collected from various locations and phosphorus contents were determined by Olsen's method ( $\text{NaHCO}_3$  extraction) and by Pauuw van der (1969) (EUF extraction) simultaneously. In another study, the soils were extracted by EUF but the color developing reagents were of Olsen's method.

A fully automatic EUF equipment, model 723 was used for the extraction of sand and soil samples. Total extraction duration lasted for 35 minutes with 4 minute intervals. The voltage between the electrodes was 50V for 0-5 min; 220V for 5-30 min; and 400V for 30-35 min according to the recommendations of Nemeth (1972, 1976 and 1979). Anode and cathode filters were of EUF-510 and EUF-511 types, respectively. The anode and cathode extracts were collected separately. The vacuum in the 2 outside chambers was adjusted so that the volume of the cathode and anode extract (soil extract + rinse water) was approximately 50 ml per extraction time.

## Results and discussion

The river sand washed with concentrated acids used in this investigation was chosen to avoid absorption and adsorption processes, and to assess desorption values of P content. The P contents were determined by means of EUF and the results obtained are presented in Table 1.

In 20 instances, P was added to sand in order to check the EUF method's reproducibility. To determine the recovery of P it was necessary to assume that all of the P present in sand was recovered by EUF. This assumption was confirmed by the recovery of 96% of the P added (Table 1).

These EUF-P values can only be attributed, when the dilution factor is taken into account by the Nemeth (1976) formula:

$$\text{Nutrient in mg/100 g} = \frac{\text{Nutrient in ppm}^* \times 50 \times 100}{5^{**} \times 1000}$$

\* = Nutrient in ppm (filtrate). 50 ml measuring flask

\*\* gram material extracted.

In the given formula, the dilution factor (aliquot) taken + colour developing reagent has been omitted.

The modified Nemeth's formula has an advantage over the original formula in that it takes into account the dilution factor.

$$\begin{aligned} \text{mg P/100g} &= \frac{\text{Nutrient in ppm} \times 50 \times 100 \times 20}{5 \times 10 \times 100} = \\ &= \text{P in ppm (filtrate)} \end{aligned}$$

Table 1

*Phosphorus contents and recovery of P either present in or added to sand as determined by Electro-ultrafiltration (EUF)*

Electrodes	Extraction classes (minutes)	EUF-P (ppm) in concentration (extracts)	
		Sand sole	Sand + 5 ppm "P" of $\text{KH}_2\text{PO}_4$ *
Anode	0- 5	0.278	0.112
	5-10	0.112	0.590
	10-15	0.112	0.578
	15-20	0.112	0.554
	20-25	0.112	0.545
	25-30	0.056	0.529
	30-35	0.056	0.612
Cathode	0- 5	0.056	0.532
	5-10	0.056	0.493
	10-15	0.000	0.339
	15-20	0.000	0.279
	20-25	0.000	0.237
	25-30	0.000	0.226
	30-35	0.000	0.123
Sum of =		0.950	5.749
Recovery = 96%			

Sd = 9.087

\* = Mean of 20 samples.

The EUF-P values calculated by Nemeth's formula are lower by 50% in calcareous soils of Pakistan (Nemeth and Makhdum, 1981). So values obtained in parts per million may be multiplied by factor 2 (aliquot : colour developing reagent 1 : 1) in the original Nemeth's formula (1976) or the modified formula may be adopted.

Obviously, the evaluation of P by the standard EUF method is very reliable and can be correlated ( $r = 0.93^{**}$  at 0-10 mm) to 0.5 M  $\text{NaHCO}_3$  extractions (Olsen et al. 1954). Loch et al. (1980) also reported correlation coefficients between EUF and Olsen method and it was  $r = 0.89^{**}$  at 0-10 min. and  $r = 0.87^{***}$  at 0-30 min. fractions.

The correlation coefficient between the Olsen's P contents and the EUF-P desorbable values within 0-10 minutes determined by Pauuw van der method was significant and the relationship could best be described by the equation:

$$Y = 0.1946x - 0.2346 \quad (r = 0.93^{**}).$$

When  $Y = \text{EUF-P}$  and  $x = \text{available-P by } 0.5 \text{ M } \text{NaHCO}_3 \text{ extraction}$ .



But there was a loose correlation ( $r = 0.41^{NS}$ ) between the Olsen's P contents and EUF-P desorbable in 0-30 min. fraction by Pauw van der method.

In the other situation, the correlation coefficient between Olsen's P contents and EUF-P desorbable values within 0-10 minutes was significant and the relationship was described by:

$$Y = 0.1247x - 0.121 \quad (r = 0.90^{**})$$

Similarly, the coefficient of correlation was also significant for 0-30 minute fractions, as given below:

$$Y = 0.325x + 1.778 \quad (r = 0.63^{**})$$

The lower correlation value between EUF-P (0-30 min.) and Olsen's phosphorus may be attributed to the fact that EUF-"P" takes into account

Table 2

*Correlation coefficient for characterization of relation of EUF and conventional method of soil investigation*

Sample No.	Olsen-P (ppm)	EUF-P (ppm)			
		Pauw van der method		Olsen method's colour developing reagents	
		Desorbable within 0-10 min.	Desorbable within 0-30 min.	Desorbable within 0-10 min.	Desorbable within 0-10 min.
1	7.00	0.84	6.56	0.60	3.60
2	8.00	1.40	9.48	0.80	5.30
3	9.00	1.68	7.81	1.00	4.40
4	8.00	1.12	5.17	0.70	3.54
5	14.00	2.12	10.48	1.50	6.90
6	6.00	0.98	7.53	0.50	4.30
7	6.00	0.84	5.86	0.60	2.20
8	6.00	0.98	7.11	0.60	2.40
9	12.00	2.10	5.74	1.20	3.00
10	14.00	2.37	16.16	1.40	5.80
11	7.00	0.98	11.93	0.60	3.70
12	6.00	0.98	10.45	0.70	3.60
13	8.00	1.26	12.12	0.68	3.58
14	8.00	0.98	6.84	0.70	4.90
15	10.00	1.68	5.56	1.20	4.70
16	6.00	1.34	5.45	0.96	4.26
17	6.00	0.97	9.33	0.69	6.69
18	11.00	2.10	7.12	1.50	5.10
19	11.00	2.24	9.49	1.60	6.80
20	14.00	2.79	11.15	2.00	8.00

various physicochemical properties of the soil, such as pH, organic matter, clay mineralogy and its content of various clay minerals and calcium carbonate, etc. (Nemeth 1979).

The popular method of determining phosphorus in Pakistan soil testing laboratories is that of Olsen et al. (1954). This provides a solitary factor in involving P in soil solution. However, this method could not establish its significance for correlating soil P and seed cotton yield (Chaudhry, 1972 and Khan 1979). One of the main disadvantages of this method is the use of chemical extractant, which dissolves the more or less phosphorus which is not available to plant roots. Moreover, the method does not take into account the clay content, type of clay, organic matter and  $\text{CaCO}_3$ . Calcium carbonate has strong influence on the availability of phosphorus (Nemeth, 1979). The dominant source of phosphorus in our soils is octacalcium phosphate (Sharif et al. 1974). Therefore, studies involving P should take into account not only immediately available P but also buffering capacity and "Pool" fertilization (Gartel, 1966 and Eifert et al., 1982). For practical plant nutrition, that method should be adopted, by which soil analysis can be significantly correlated with crop yield (Wiklicky, 1982). The EUF procedure offers this choice to adopt in Pakistan (Malik et al. 1984).

The electro-ultrafiltration (EUF) by employing varying voltages and temperature and using water as a very mild extractant has, gained superiority over Olsen's method (Wiklicky, 1982). This provides a series of data on P dynamics in soil (Nemeth and Makhdum, 1981). This method includes all influencing factors (Nemeth, 1972) and provides data on:

- (a) concentration in soil solution (0–10 min. fractions);
- (b) buffering capacity (10–30 min. fractions);
- (c) reserve nutrient (30–35 min. fractions);

### Conclusions

The reproducibility of EUF-P in the different fraction can best be correlated with Olsen-P when the dilution factor is considered and evaluated. Moreover, independent of the amount of phosphorus either present or added to the soil, the present modified formula gave in all cases highly replicable quantitative data.

Another important feature of the EUF is that it determines quantitatively such nutrient dynamics as N, P, K, Na, Ca and heavy metals in soil as well as characteristic soil properties such as clay content, clay mineralogy, organic matter and  $\text{CaCO}_3$ , etc., by employing simultaneously varying voltage and temperature, this unique opportunity cannot be provided by any other conventional methods currently used in soil laboratories.



## References

- Chaudhry, T. M. (1972): Cotton soils of Pakistan. In *Cotton in Pakistan*. Pakistan Central Cotton Committee, Karachi. pp. 287—294.
- Eifert, J., M., Várnai, L., Szőke (1982): Application of the EUF procedure in grape production. *Plant and soil*. **64** (1), 105–113.
- Gartel, W. (1966): Über die Düngung der Reben in intensive bewirtschafteten Weinbaugebieten. *Weinbau und Keller*, **7**, 295–336.
- Khan, M. A. (1979): Phosphorus nutrition of the cotton plant. *The Pak. Cottons* **23**, 147–169.
- Loch, J., Kiss, Sz., Jászberényi, I., Vágó, I. (1980): *Relation between the nutrient content of soil and uptake of PK, Ca, Mg by perennial ryegrass*. Proc. of Internal. Symp. on the "Application of Electro-ultrafiltration in the Agricultural Production", held at Budapest (Hungary) on 6–10 May, pp. 207–215.
- Malik, M. N., Makhdum, M. I., Chaudhry, F. I. (1984): *Application of electro-ultrafiltration (EUF) for predicting phosphorus availability to cotton*. Proc. of II. Internal. Symp. on the "Application of Electro-ultrafiltration in the Agricultural Production", held at Vienna on 5–10 May.
- Nemeth, K. (1972): *The determination of desorption and solubility rates of nutrients in the soil by means of electro-ultrafiltration (EUF)*. Proc. 9th Colloq. International Potash Institute, pp. 171–180.
- Nemeth, K. (1976): The effective and potential availability of nutrients in the soil by means of electro-ultrafiltration (EUF). *Appl. Sci. Develop.* **8**, 89–111.
- Nemeth, K. (1979): The availability of nutrients in the soil as determined by electro-ultrafiltration (EUF). *Adv. Agron.* **31**, 155–186.
- Nemeth, K., Makhdum, M. I. (1981): Evaluation of the nutrient dynamics in calcareous soils from Pakistan by electro-ultrafiltration (EUF). *Soil Sci. Plant Nutr.* **27** (2), 159–168.
- Olsen, S. R., Cole, C. V., Wantabe, F. S., Dean L. A. (1954): Estimation of available phosphorus in soils by extraction with sodium bicarbonate. *US Dept. Agric. Cir.* **939**, 1–19.
- Pauuw, F. van der (1969): Development and evaluation of new water extraction technique for determination of available phosphate. *Landw. Forsch.* **23**, 102–109.
- Sharif, M., Chaudhry, F. M., Lakho, A. G. (1974): Suppression of superphosphate phosphorus fixation by farmyard manure II. Some studies on the mechanisms. *Soil. Sci. Plant Nutr.* **20**, 395–401.
- Wiklicky, L. (1982): Application of the EUF procedure in sugar beet cultivation. *Plant and Soil*. **64** (1), 115–127.



## DISTRIBUTION OF NATIVE FIXED AMMONIUM AND ITS PREDICTION AS BASED ON TEXTURAL COMPONENTS AND ORGANIC CARBON IN SOME SUBTROPICAL SOILS OF WEST BENGAL AND BIHAR (INDIA)

A. DAS and B. DATTA

SOIL SCIENCE SECTION, AGRICULTURAL ENGINEERING DEPARTMENT,  
I. I. T., KHARAGPUR, INDIA

(Received: 23 June 1987; accepted 1 September 1987)

Native fixed ammonium ( $\text{NH}_4\text{-N}$ ) and carbon-nitrogen ratio (C/N) of nine subtropical soil profiles, of normal to moderate depth, representing different soil types and sampling localities in the state of West Bengal and Bihar (India) were studied. The results obtained were discussed and utilized in determining simple correlation coefficients between fixed  $\text{NH}_4\text{-N}$  and different soil characteristics (pH, CEC, organic carbon and textural components) separately. For each soil a particular soil parameter based on the highest value of "coefficient of determination" ( $r^2$ ) was identified and a linear relationship of that with fixed  $\text{NH}_4\text{-N}$  was calculated for predicting the percentage distribution of fixed  $\text{NH}_4\text{-N}$ . Some of the salient findings were as follows:

(1) Irrespective of soils the fixed  $\text{NH}_4\text{-N}$  as percent of total N was found to vary from 18.2 to 71.7 in an individual horizon.

(2) Variation in the content of fixed  $\text{NH}_4\text{-N}$  amongst different series was not appreciable. However, Oxisols (Garbeta, Bagru), Ultisol (Lal Gutuwa) and the soils of more gravelly to sandy nature contained a lower percentage of fixed  $\text{NH}_4\text{-N}$ .

(3) Within a profile, lower horizons often contained a higher percentage of fixed  $\text{NH}_4\text{-N}$ .

(4) C/N ratio almost in all the soils was found to decrease with depth, and the narrowing of C/N ratio was attributed largely to fixed  $\text{NH}_4\text{-N}$ .

(5) That a major part of fixed  $\text{NH}_4\text{-N}$  was associated with the finer textural fractions of the soils was evident from a positive correlation of it with fine clay ( $r = 0.09$  to 0.95), clay ( $r = 0.50$  to 0.97) and clay + silt ( $r = 0.22$  to 0.97).

(6) Except for Kharagpur soil (ultic Haplustalf) fixed  $\text{NH}_4\text{-N}$  showed a positive correlation ( $r = 0.15$  to 0.95) with CEC of soils.

(7) For the prediction of fixed  $\text{NH}_4\text{-N}$ , "sand percentage" (whole soil basis) was found to be the particular parameter governing those linear regressions which could account for 98%, 77%, 93% and 98% variation of the total fixed  $\text{NH}_4\text{-N}$  respectively for Kharagpur (Ultic Haplustalf), Labani (Rhodustalf), Lal Gutuwa (Oxic Rhodustalf) and Bagru (Ultic Gissburg) soils; while "clay", "fine clay", "silt + clay" (all based on whole soil) and organic carbon percentage respectively for Garbeta (Ultic Acrustox), Kurchiboni (Oxic Rhodustalf), Silda (Chromustert) and both Bara Ara (Anda Eutropept) and Netarhat (Vertic Eutropept) soils were the controlling parameters of those linear regressions which would govern 93%, 71%, 99%, 56% and 58% variations of the total fixed  $\text{NH}_4\text{-N}$  for the above soils in chronological order.

**Keywords:** ammonium, organic carbon, soil texture, subtropical soils



## Introduction

High temperature and rainfall in the tropics and subtropics are responsible for accelerated oxidation and/or decomposition of soil organic matter, transformation of soil nitrogen and its increased loss by leaching and volatilization. Under such agroclimatic and soil condition, fixation of ammonium nitrogen ( $\text{NH}_4\text{-N}$ ) in exchangeable form by layer silicate minerals or hydrous oxides and organic matter is very likely to contribute to the nitrogen nutrition and economy of these soils. Many investigators in the past having used different extractants for the evaluation of fixed  $\text{NH}_4\text{-N}$  and reported that fixed  $\text{NH}_4\text{-N}$  would make up a substantial portion of soil nitrogen especially in the subsoils. Rodrigues's (1954) data for four Caribbean tropical soils were found to be high and maintained a mixed trend with depth, while results reported by Dhariwal and Stevenson (1958) showed an increase of fixed  $\text{NH}_4\text{-N}$  from surface to subsoils (4% to 78% total N). Similarly, the results obtained by Moore and Ayeke (1965) with Nigerian soils, which showed a greater native fixed  $\text{NH}_4\text{-N}$  in the subsoil than in the surface, were contrary to that of Mikama and Kanehiro (1968) where a general decrease of fixed N had been depicted with depth and the values of which ranged from 0 to 585 ppm (0 to 32.9 as percent of total N) in an individual soil horizon of Hawaii. Martin et al. (1970) discovered that fixed  $\text{NH}_4\text{-N}$  increased and attained very high values in each subsoil (one contained 0.108% N) as total N contents decreased with depth. Opuwaribo and Odu (1974), and Dalal (1977), after studying Nigerian and Trinidad soils, respectively reported that the fixed  $\text{NH}_4\text{-N}$  had accounted for 1% to 7% of total N in surface and 3% to 48% in subsoils of Nigeria, and as high as 77% in the subsoils of Trinidad. Sah and Pasricha (1984) investigating some soils of Punjab (India) described that 22% to 87% of total N was present as fixed  $\text{NH}_4\text{-N}$  a higher percentage of the latter being invariably observed at lower depth. He also showed fixed  $\text{NH}_4\text{-N}$  to be associated with the finer textural fractions of soils. Major causes which led to the high values of fixed  $\text{NH}_4\text{-N}$  in tropical and subtropical soils were (1) the use of strong HCl-HF acid as extractant which would cause a degradation of organic nitrogen (Freney 1964; Bremner 1967) and (2) the estimation of nitrogen by Kjeldahl procedure which could not recover all the nitrogen present as fixed  $\text{NH}_4\text{-N}$  (Stewart and Porter 1963; Tewari et al. 1969).

As little information is available on the distribution of fixed  $\text{NH}_4\text{-N}$  as well as its relation to soil characteristics in the subtropical soils of West Bengal and Bihar (India), and attempt was made to study this and to determine for each soil a simple linear regression for predicting the percentage distribution of fixed  $\text{NH}_4\text{-N}$  to a greater extent, by any of the ordinary soil characteristics, in order to utilize those for all practical purposes so as to avoid the tedious processes involved in the continual estimation of fixed  $\text{NH}_4\text{-N}$ .



## Materials and methods

Soil samples and samples from the parent material and geological layers below that of nine subtropical soil series, namely Kharagpur (Ultic Haplustalf), Garbeta (Ultic Acrustox), Labani (Rhodustalf), Kurchiboni (Oxic Rhodustalf), Silda (Chromustert), Bara Ara (Anda Eutropept), Lal Gutuwa (Oxic Rhodustalf), Baguru (Ultic Gissburgiorthox), and Netarhat (Vertic Eutropept) occurring in the states of West Bengal and Bihar (India) were used in the present study. Particle size distribution (expressed on whole soil basis) and other physicochemical properties of soils were determined from the sieved sample ( $<2$  mm) following the standard methods as described by Back (1965) and Jackson (1973), and are presented in Table 1. Native fixed  $\text{NH}_4\text{-N}$  was determined by the method developed by Silva and Bremner (1966) wherein one gram ground (100 mesh) sample was treated with 20 ml alkaline KOBr to remove exchangeable and organic nitrogen compounds. The residue was washed several times with 0.5 N KCl solution and then shaken continuously for 24 hours with 20 ml of 0.5 N HF-1N HCl solution. The ammonium released by this treatment was steam distilled with 10 N NaOH into a boric acid-methyl red-bromo cresol green indicator solution, by means of a *micro-Kjeldahl* steam distillation apparatus, and finally titrated with 0.005 N  $\text{H}_2\text{SO}_4$ . Total nitrogen (N) was estimated by *Kjeldahl* procedure and exchangeable  $\text{NH}_4\text{-N}$  and  $\text{NO}_3\text{-N}$  were determined by extraction of 20 g soil with 10 ml 1N KCl solution in a 200 ml erlenmeyer as given by Cottenie (1980). Organic carbon was determined by the Walkley and Balck method (Balck 1965). The difference between total N and inorganic N (fixed  $\text{NH}_4\text{-N}$  + exchangeable  $\text{NH}_4\text{-N}$  and  $\text{NO}_3\text{-N}$ ) was taken as the value of organic N.

## Results and discussion

The data presented in Table 1. indicate that most of the soils contained a high percentage of sand or gravel and for this reason the samples involved in the present study were collected to a moderate depth (normal) with a view to extend the results obtained even over the area of deep-rooted or plantation crops management practices, in case any of these soils were otherwise found unsuitable for general cropping practices.

As is evident from Table 2, an appreciable amount of fixed  $\text{NH}_4\text{-N}$  both as percentage of total N and in absolute term was present in the soils and the values of which ranged from 18.2% to 71.2% and 38.8 to 478.5 ppm (data not shown, but could be computed from Table 2) respectively. Martin et al. (1970) for subtropical soils had also reported such high values for the fraction of fixed  $\text{NH}_4\text{-N}$ . The soils did not show any definite pattern in the profile as to the relative distribution of fixed  $\text{NH}_4\text{-N}$  in an absolute term (not shown in Table 2). However, a slight decreasing trend in some of the profiles depended mostly on the coupled effect of total N as well as the content and type of clays. When fixed  $\text{NH}_4\text{-N}$ , as a percentage of total N was considered, it was noticed that three soils (Garbeta, Labani and Silda) clearly showed a mixed trend with depth, while that in rest of the soils tended to increase, although the highest value was not always associated with the lowest horizon (Table 2). This percentage increase of fixed  $\text{NH}_4\text{-N}$  (as fraction of total N) with depth was due either to (1) the decrease of total N down the profile or (2) the increase in the absolute content of fixed  $\text{NH}_4\text{-N}$  arising out of higher content of illite and montmorillonite clays or (3) both the factors (1) and (2),



**Table 1**  
*Some physico-chemical*

Soil series	Classification	Clay minerals <sup>1</sup>	Depth (cm)	pH
1	2	3	4	5
Kharagpur	Ultic Haplustalf	I, K (major) M (minor)	0- 15	5.20
			15- 80	4.60
			80-130	4.80
			130-195	5.20
			195-225	5.30
Garbeta	Ultic Acrustox	K (dominant), I	0- 20	5.10
			20-122	4.10
			122-165	5.70
			165-175	5.60
			175-217	5.50
			217-227	5.80
			227-320	5.10
			320-427	5.50
			427-477	4.90
			477-500	5.10
Labani	Rhodustalf	M, K & I in varying propor- tions with depth	0- 15	5.20
			15- 35	5.70
			35- 95	5.65
			95-150	5.70
			150-215	6.30
Kurchibani	Oxic Rhodustalf	K, M and/or I in varying proportions with depth.	0- 15	4.20
			15- 25	5.90
			25- 75	6.30
			75-115	6.50
			115-140	7.10
Silda	Chromustert	M (dominant), I, K	140-210	8.30
			0- 80	5.40
			80-125	7.35
			125-210	8.00
			210-365	6.00
Bara Ara	Andic Eutropept	M (upper hori- zons) or V (lower hori- zons) dominant, K (minor)	365-	6.90
			0- 10	6.60
			10-125	6.20
			125-200	5.70
			200-295	6.20
Lal Gutuwa	Oxic Rhodustalf	K (dominant) M (trace)	295-425	6.10
			425-	6.50
			0- 20	5.70
			20-130	5.40
			130-190	5.25
Bagru	Ultic Gibbsi- orthox	K, I (upper horizons); K, G (lower horizons)	190-320	5.1
			320-	6.6
			0- 50	5.50
			50-100	5.80
			150-210	5.50
			210-	5.80

*properties of soils*

CEC meq/100 gm	Organic carbon (%)	Partiolo-size <sup>2</sup> distribution (%)				
		Gravel	Sand	Silt	Coarse clay	Fine clay
6	7	8	9	10	11	12
12.2	0.34	nil	61.5	13.1	17.9	7.2
9.0	0.14	nil	55.7	12.2	22.4	9.6
9.3	0.12	nil	53.6	11.0	23.3	11.8
8.1	0.10	0.50	48.0	12.1	24.8	15.5
7.8	0.11	1.50	48.1	12.5	25.5	12.0
7.8	0.32	50.5	34.91	4.1	6.9	3.3
5.8	0.25	45.8	39.4	3.2	7.8	3.7
7.3	0.30	20.5	43.5	8.0	16.5	11.5
5.2	0.23	32.4	49.6	5.2	9.6	3.6
6.5	0.23	28.8	39.2	4.2	16.8	10.3
5.3	0.21	35.8	50.1	3.0	8.4	2.6
5.1	0.20	9.65	34.8	17.1	21.9	16.6
4.9	0.19	15.3	51.4	13.1	13.2	7.1
3.4	0.18	19.9	50.7	7.3	14.1	7.9
8.8	0.30	25.0	53.7	8.9	9.1	3.2
11.5	0.31	0.5	7.1	43.8	25.7	22.8
12.3	0.31	1.0	10.9	37.4	26.5	24.0
8.0	0.78	37.5	28.0	20.2	11.0	2.9
8.5	0.42	41.23	31.3	11.0	12.5	3.7
9.2	0.33	43.5	15.8	13.3	19.9	7.4
10.0	0.32	42.5	19.4	12.5	16.8	8.7
10.5	0.26	26.8	30.0	28.1	11.9	3.0
11.2	0.90	13.4	33.3	29.2	17.3	6.8
10.0	0.36	48.4	21.0	7.1	18.7	4.4
11.3	0.27	57.0	3.5	14.3	19.8	5.3
10.2	0.21	63.8	8.9	5.1	16.3	5.7
12.8	0.19	61.6	7.2	11.5	14.3	5.2
17.9	0.18	4.4	38.0	24.0	20.9	12.5
19.0	0.66	11.8	42.0	23.9	17.2	4.9
38.0	0.33	9.9	18.6	24.0	30.8	16.5
42.5	0.30	2.7	29.2	19.5	29.6	18.8
18.1	0.23	42.2	25.5	6.1	16.7	9.3
45.2	0.17	6.0	8.2	33.5	30.3	21.9
20.1	0.69	43.0	20.4	16.2	13.2	7.0
33.0	0.36	34.7	15.1	23.5	15.3	11.2
37.5	0.18	20.3	25.2	15.5	16.1	22.4
48.4	0.15	13.4	21.0	31.5	16.8	17.2
40.5	0.12	19.0	22.1	27.1	19.9	12.3
42.2	0.10	21.2	20.8	30.5	15.8	11.2
4.3	0.94	70.2	4.40	11.9	10.0	3.5
3.2	0.74	66.2	10.30	8.1	9.9	5.3
3.0	0.20	50.5	23.2	8.0	10.0	8.0
5.6	0.08	0.5	39.3	20.3	21.7	17.8
5.8	0.09	nil	45.2	26.2	12.7	13.3
5.2	1.34	3.20	43.10	27.2	21.3	5.0
6.3	0.82	nil	23.7	26.3	37.4	12.5
6.1	0.38	nil	22.1	29.9	29.4	18.4
4.2	0.19	10.0	50.80	12.1	21.4	5.7



Table 1 (contd.)

Soil series	Classification	Clay minerals <sup>1</sup>	Depth (cm)	pH
1	2	3	4	5
Netarhat	Vertic Eutropept	M (dominant), V, K	0- 30	6.40
			30- 40	6.45
			48- 60	6.15
			60- 90	6.20
			90-120	6.00
			120-145	6.00
			145-176	5.80
			176-	6.50

\* The soil layers as well as geological layers are investigated and the results evaluated together. — That of course is to consider by the consequences based on the results.

<sup>1</sup> Mineral designations: M = Montmorillonite; I = Illite; V = Vermiculite; K = Kaolinite; G = Gibbsite.

while the mixed trend of that in some of the profiles could be explained by a sudden increase in total N or a sudden decrease in the absolute amount of fixed  $\text{NH}_4\text{-N}$  or both. Thus, it could be inferred that illite, montmorillonite and/or vermiculite clays that most of the soils (except for Lal Gutuwa) contained (Table 1) also played a major role, besides the content of total N being a cause, in deciding the extent of fixed  $\text{NH}_4\text{-N}$  both as percent of total N and in absolute terms.

The organic N ranged from 19.4% to 78.2% of total N with a general tendency to decline (with a few exceptions) down any profile, although the smallest fraction for each profile was not necessarily associated with the lowermost horizon (Table 2). It is also evident (Table 2) that the pattern of distribution of organic N fraction was exactly opposite to that of fixed  $\text{NH}_4\text{-N}$  i.e., an upward trend in organic N fraction from the bottom towards the top of a profile (except for few profiles e.g., Garbeta, Labani, etc. for which the trends were of a somewhat mixed type). Any sharp abatement of organic N with depth was mostly due to an decrease in its absolute amount, while the narrowing of organic N/total N ratio with depth or a mixed trend of the same (Table 2), in general, came into effect as a result of a varied distribution of fixed  $\text{NH}_4\text{-N}$  in the soil. Moore and Ayeke (1965) opined similarly, for Nigerian soils, that the declining trend of organic N/total N ratio with depth was attributed to fixed  $\text{NH}_4\text{-N}$ . Organic C/organic N ratios in general varied within a very narrow range in most of the soils. However, in some profiles a considerable increasing or decreasing trend of the ratio had also been observed (Table 2). Some of the lower values of the ratio in different profiles, or within a profile at lower depths, were thought to have occurred due to a low recovery of fixed  $\text{NH}_4\text{-N}$ , as stated by Dalal (1977), as well as for having in the soil a relatively



CEC meq/100 gm	Organic carbon (%)	Particle-size <sup>2</sup> distribution (%)				
		Gravel	Sand	Silt	Coarse clay	Fine clay
6	7	8	9	10	11	12
32.2	1.58	0.5	41.8	29.5	21.5	6.5
23.0	0.72	nil	50.2	31.0	14.2	3.8
36.8	0.38	nil	10.5	59.0	21.4	8.8
39.0	0.28	nil	44.0	21.3	23.5	10.8
20.4	0.22	nil	40.8	32.8	19.8	6.2
33.3	0.20	nil	30.6	40.3	22.0	6.5
37.5	0.18	nil	25.0	42.0	24.0	8.0
18.5	0.18	nil	52.2	22.1	19.3	6.0

<sup>2</sup> Particle size: Gravel = <2 mm; Sand = (2-0.05) mm; Silt = (0.05-0.002) mm; Coarse clay = (0.002-0.0002) mm; Fine clay = <0.0002 mm. Total clay = Fine clay + coarse clay

higher among of finer fraction and oxides/hydroxides of iron and aluminium. The reason for some exceptional high values of the ratio in different soils or within a profile with increasing depth, such as Netarhat, was uncertain and the fact contradicted our expectations. However, a possible explanation for this, though it seems very remote, might be the overestimation of fixed  $\text{NH}_4\text{-N}$  which resulted in the decrease of organic N and thereby caused an increment of the ratio. An excess amount of ammonium from soils treated with Silva and Bremner (1966) extractant possibly resulted from the degradation of low molecular organic nitrogen compound remained nonoxidised for having been masked by hydroxides of iron or sorbed in the interlamellar space of the montmorillonite and/or vermiculite clay that the Netarhat and Bara Ara soils contained (Table 1).

Table 3 represents the coefficients of correlation ( $r$ ) between  $\text{NH}_4\text{-N}$  and textural fractions, organic carbon, CEC and pH of soils. It is evident from Table 3 that the clay content of the soils was positively correlated ( $r = 0.56$  to  $0.97$ ) with fixed  $\text{NH}_4\text{-N}$ , showing significant relationship only for Kharagpur ( $r = 0.97$ ), Garbeta ( $r = 0.96$ ) and Silda ( $r = 0.94$ ) soils, while the fine clay content with the fixed  $\text{NH}_4\text{-N}$  depicting positive correlation ( $r = 0.09$  to  $0.95$ ) showed, besides Kharagpur ( $r = 0.91$ ), Garbeta ( $r = 0.95$ ) and Silda ( $r = 0.89$ ) soils, a significant relationship also for Kurchiboni ( $r = 0.84$ ) soil. That the Lal Gutuwa soil profile exhibited a very poor positive correlation ( $r = 0.09$ ) between fixed  $\text{NH}_4\text{-N}$  and fine clay (Table 3) was indicative of practically no relation between the two variables. Similarly, the value of  $r = 0.03$  for Netarhat soil between fixed  $\text{NH}_4\text{-N}$  and silt, hardly indicated and association. The value  $r$  was found to range from  $0.03$  to  $0.90$  in the soils (except for Kharagpur soil which came out with an  $r$  value of  $-0.42$ ) when



Table 2

Total N percentage distribution of fixed  $\text{NH}_4\text{-N}$ , organic N and C/N ratio of soil profiles

Depth (cm)	Organic carbon (%)	Total nitrogen (%)	C : N (total) ratio	Fixed $\text{NH}_4\text{-N}$ (% of total)	Organic N (% of total)	C : N (organic) ratio
(1)	(2)	(3)	(4)	(5)	(6)	(7)
Kharagpur (Ultic Haplustalf)						
0-15	0.34	0.028	12.14	26.6	65.1	18.65
15-80	0.14	0.012	11.67	32.3	51.9	22.48
80-130	0.12	0.012	10.00	37.8	48.0	20.83
130-195	0.10	0.013	7.69	42.9	45.1	17.66
195-225	0.11	0.015	7.33	43.2	47.3	15.50
Garbeta (Ultic Acrustox)						
0-20	0.32	0.029	11.03	18.2	78.2	14.10
20-122	0.25	0.024	10.42	18.6	77.0	13.53
122-165	0.30	0.031	9.67	34.5	61.8	15.64
165-175	0.23	0.023	10.00	30.2	64.6	15.25
175-217	0.23	0.028	8.21	36.3	60.1	13.67
217-227	0.21	0.026	8.08	26.2	70.3	11.49
227-320	0.20	0.028	7.14	44.8	51.1	13.97
320-427	0.19	0.027	7.04	35.2	60.9	11.55
427-477	0.18	0.029	6.20	38.0	58.8	10.54
477-500	0.30	0.043	6.98	25.1	72.2	9.67
500-595	0.31	0.053	5.85	59.3	38.5	15.17
595-642	0.31	0.054	5.74	63.5	33.8	16.99
Labani (Rhodustalf)						
0-15	0.78	0.080	9.75	54.3	40.6	23.99
15-35	0.42	0.038	11.05	39.2	53.0	20.86
35-95	0.33	0.037	8.92	64.3	28.2	31.63
95-150	0.32	0.041	7.80	60.8	34.7	22.51
150-215	0.26	0.034	7.65	52.2	43.4	17.62
Kurchibani (Oxic Rhodustalf)						
0-15	0.90	0.081	11.11	59.2	35.5	31.33
15-25	0.36	0.041	8.78	42.3	49.9	17.60
25-75	0.27	0.032	8.43	52.1	39.4	21.42
75-115	0.21	0.030	7.0	52.2	40.8	17.16
115-140	0.19	0.029	6.20	56.5	38.2	16.25
140-210	0.18	0.025	7.20	66.8	28.1	25.62
Silda (Chromustert)						
0-80	0.66	0.058	11.38	48.3	44.4	25.63
80-125	0.33	0.024	13.75	64.5	27.3	50.35
125-210	0.30	0.022	13.63	60.2	31.3	43.49
210-365	0.23	0.028	8.21	42.7	50.69	16.20
365+	0.17	0.022	7.72	69.7	25.2	30.65
Bara Ara (Andic Eutropept)						
0-10	0.690	0.070	9.85	45.2	48.0	20.52
10-125	0.360	0.035	10.29	38.4	53.3	19.29
125-200	0.180	0.019	9.47	55.7	34.0	27.85
200-295	0.15	0.018	8.33	59.4	31.4	26.50
295-425	0.12	0.014	8.57	58.3	32.8	26.16
425+	0.10	0.013	7.69	63.9	26.5	28.97

Table 2 (contd.)

Depth (cm)	Organic carbon (%)	Total nitrogen (%)	C : N (total) ratio	Fixed $\text{NH}_4\text{-N}$ (% of total)	Organic N (% of total)	C : N (organic) ratio
(1)	(2)	(3)	(4)	(5)	(6)	(7)
Lal Gutuwa (Oxic Rhodustalf)						
0- 20	0.94	0.081	11.60	19.2	74.0	15.67
20-130	0.70	0.062	11.29	28.3	66.7	16.92
130-190	0.20	0.025	8.0	32.3	57.0	14.03
190-320	0.08	0.011	7.27	49.2	38.0	19.13
320-430	0.09	0.020	4.5	65.4	29.1	15.46
Bagru (Ultic Gibbsiorthox)						
0- 50	1.34	0.092	14.57	37.2	56.7	25.71
50-150	0.82	0.085	10.37	46.2	49.7	20.88
150-210	0.38	0.045	8.44	49.7	44.4	19.00
0- 50	1.34	0.092	14.57	37.2	56.7	25.71
50-150	0.82	0.085	10.37	46.2	49.7	20.88
150-210	0.38	0.045	8.44	49.7	44.4	19.00
210-	0.19	0.028	6.78	32.3	61.6	11.01
Netarhat (Vertic Entropept)						
0- 30	1.58	0.102	15.49	34.2	59.9	25.81
30- 48	0.72	0.052	13.85	30.3	62.1	22.30
48- 60	0.38	0.035	10.85	53.7	35.9	30.20
60- 90	0.28	0.027	10.37	58.9	28.8	35.96
90-120	0.22	0.018	12.22	48.8	36.6	33.37
120-145	0.20	0.018	11.11	59.0	28.3	39.30
145-176	0.18	0.015	12.0	67.6	19.4	61.85
176-	0.18	0.016	11.25	71.7	29.6	57.45

Results based on average of three measurements.

the relationship between fixed  $\text{NH}_4\text{-N}$  and silt content was drawn. However, the evidence of above-mentioned association for Kharagpur soil did not prove that a decrease in fixed  $\text{NH}_4\text{-N}$ . The evidence of such causality must come from other sources. When clay in combination with silt was considered together, the relationship with fixed  $\text{NH}_4\text{-N}$  was significant for four soil series i.e., Kharagpur ( $r = 0.97$ ), Garbeta ( $r = 0.95$ ), Silda ( $r = 0.99$ ) and Bagru ( $r = 0.99$ ). Except for Kurchibani and Lal Gutuwa soils, the relationship between fixed  $\text{NH}_4\text{-N}$  and sand fraction was inverse ( $r = -0.10$  to  $-0.99$ ). That the fixed  $\text{NH}_4\text{-N}$  exhibited a negative relationship with sand and a positive relationship with finer fractions (i.e. clay, fine clay and clay + silt) suggested that the finer fractions were the most responsible inorganic components for the bulk of  $\text{NH}_4\text{-N}$  fixation. The suggestion was supported by the evidence that the finer fraction (considering only the clay) of the soils under study were rich in illite and/or montmorillonite and/or vermiculite clay which were reported by many investigators to be highly susceptible for ammonium fixation. Convincing evidence of association for Kurchibani ( $r = 0.53$ ) and Lal



**Table 3**  
Coefficient of correlation between fixed  $\text{NH}_4\text{-N}$

Soil series	Value of coefficient of correlation (r)				
	Sand	Silt	Clay	Silt + clay	Fine clay
Kharagpur	-0.99**	-0.42	0.97**	0.97**	0.91**
Garbeta	-0.75	0.90***	0.96***	0.95***	0.95***
Labani	-0.88*	0.00	0.72	0.74	0.69
Kurchiboni	0.53	0.74	0.61	0.61	0.84*
Silda	-0.71	0.81	0.94*	0.99***	0.89*
Bara Ara	-0.10	0.39	0.50	0.70	0.38
Lal Gutuwa	0.96**	0.90*	0.76	0.87	0.09
Bagru	-0.99**	0.81	0.93	0.99**	0.94
Netarhat	-0.20	0.03	0.56	0.22	0.47
Surface Samples (all series)	-0.03	0.69*	0.31	0.56	0.24
All samples (all series)	-0.20	0.58**	0.54**	0.66***	0.25

\*, \*\*, \*\*\* significant at the 5%, 1% and 0.1% level respectively.

Gutuwa ( $r = 0.96$ ) between fixed  $\text{NH}_4\text{-N}$  and sand percentage, as stated earlier, does not mean that the increase in sand content was the cause of the increase in fixed  $\text{NH}_4\text{-N}$  for these soils. The anomaly could be explained by looking at the declined percentage of gravel with the concomitant increase in sand and finer components down the profile (e.g., Lal Gutuwa, Table 1) or which the latter became the only factor, though the sand percentage also had an upward trend, responsible for an increase in fixed  $\text{NH}_4\text{-N}$  with depth.

Organic carbon, in general, was negatively correlated ( $r = -0.01$  to  $-0.76$ ) (with a few exceptions) with  $\text{NH}_4\text{-N}$ , suggesting that the latter was mostly associated with the inorganic components (Table 3). However, the negligible positive correlation or no relationship seen in case of Garbeta, Laboni, Kurchiboni and Bagru soil was indicative of a possible contribution, although to a little extent, from an organic component of soil towards the fixed  $\text{NH}_4\text{-N}$ .

Because of the fact that  $\text{NH}_4\text{-N}$  is fixed mostly by layer silicates through the process of cation exchange property, it is generally expected that fixed  $\text{NH}_4\text{-N}$  in soil will have some relationship with cation exchange capacity (CEC) of soil. Most of the soils under study were found to show positive correlations between the above two parameters. However, a significant relationship was found for Garbeta ( $r = 0.66$ ), Kurchiboni ( $r = 0.83$ ) and Silda ( $r = 0.95$ ) soil, while Laboni, Bara Ara and Netarhat soil showed a very poor value of (Table 3).



and textural fraction, organic carbon, CEC and pH of soils

of fixed $\text{NH}_4\text{-N}$ versus			Linear regression <sup>1</sup>
Organic C	CEC	pH	
-0.86	-0.41	0.32	$Y = 103.04 - 1.25x$ (sand %); ( $r^2 = 0.98$ )
0.14	0.66*	-0.01	$Y = 12.65 + 0.95x$ (clay %); ( $r^2 = 0.93$ )
-0.15	0.33	-0.12	$Y = 84.90 - 1.23x$ (sand %); ( $r^2 = 0.77$ )
0.05	0.83*	0.37	$Y = 39.39 + 2.33x$ (fine clay %); ( $r^2 = 0.71$ )
-0.42	0.95*	0.70	$Y = 25.26 + 0.53x$ (silt + clay %); ( $r^2 = 0.99$ )
-0.75	0.31	-0.12	$Y = 61.98 - 31.86x$ (org. C%); ( $r^2 = 0.56$ )
-0.83	0.81	0.52	$Y = 14.38 + 1.00x$ (sand %); ( $r^2 = 0.93$ )
-0.01	0.57	0.62	$Y = 60.81 - 0.56x$ (sand %); ( $r^2 = 0.98$ )
-0.76*	0.15	-0.44	$Y = 63.83 - 23.11x$ (org. C%); ( $r^2 = 0.58$ )
0.17	0.19	-0.32	$Y = 15.46 + 1.19x$ (silt %); ( $r^2 = 0.48$ )
-0.25	0.43**	0.14	$Y = 32.91 + 0.70x$ (silt + clay %); ( $r^2 = 0.44$ )

Linear regression<sup>1</sup>: Between fixed  $\text{NH}_4\text{-N}$  (Y) and the "parameter" (x, as shown in the parenthesis in respective cases) showing maximum  $r$  value.

While the pH of five soils (Kharagpur, Kurchiboni, Silda, Lal Gutuwa and Bagru) showed positive correlation with fixed  $\text{NH}_4\text{-N}$ , indicating a tendency of the latter to increase with rise in pH, the other three soils (Laboni, Garbeta and Bara Ara) virtually developed no relationship between the two variables (Table 3). Sah and Pasricha (1984) obtained a positive correlation between fixed  $\text{NH}_4\text{-N}$  and pH for the soils of Punjab. The downward trend in fixed  $\text{NH}_4\text{-N}$  in presence of exchangeable  $\text{H}^+$  ions, as explained by Barshad (1954) might be due to the expanded state of crystal lattice, wherein the adsorbed  $\text{H}^+$  ( $\text{H}_3\text{O}^+$ ) ion would leave the crystal lattice, keeping inter-lattice  $\text{NH}_4\text{-N}$  accessible to a replacing cation.

When the correlation was established between fixed  $\text{NH}_4\text{-N}$  and several soil characteristics for the surface soils of all the profiles, it was observed that the former made a significant relationship only with silt (Table 3). Irrespective of localities and type of soils when all the profile samples were considered, it was noticed that the fixed  $\text{NH}_4\text{-N}$  significantly related with finer fractions i.e., with clay ( $r = 0.54$ ), silt ( $r = 0.58$ ) and silt + clay ( $r = 0.66$ ) content, which indicated a clear upward trend of the former with the increase in either clay or silt, or silt + clay, content of the soils (Table 3). The the soils in general showed a tendency to increase the fixed  $\text{NH}_4\text{-N}$  with an increase in CEC of soils ( $r = 0.43$ , Table 3) further supported the above view, as an increase in CEC possibly occurred from the increase in clay, or silt + clay, content of these soils.



From Table 3 it is obvious that amongst all the soil characteristics, "sand percentage" showed the highest value of coefficient of determination ( $r^2$ ) for Kharagpur, Laboni, Lal Gutuwa and Bagru soil, while clay, fine clay, silt + clay (all based on whole soil) and organic carbon percentages were the characteristics depicting the highest value of  $r^2$  respectively for Garbeta, Kurhiboni, Silda and both Bara Ara and Netarhat soils. Linear regressions for each soil for the prediction of fixed  $\text{NH}_4\text{-N}$  are presented in Table 3. It is evident that for Kharagpur (Ultic Haplustalf), Laboni (Rhodustalf), Lal Gutuwa (Oxid Rhodustalf) and Bagru (Ultic Gibbsiorthox) soils, the predicting equations were governed by the variation of sand content and would account for 98%, 77%, 93% and 98% variation of the total fixed  $\text{NH}_4\text{-N}$  respectively. Those for Garbeta (Ultic Acrustox), Kurchiboni (Oxic Rhodustalf), Silda (Chromustert), Bara Ara (Andic Eutropept) and Netarhat (Vertic Eutropept), soil 1 were controlled respectively by the variation of clay, fine clay, silt + clay (whole soil basis) and organic carbon percentages to account for 93%, 71%, 99%, 56% and 58% variation of the total fixed  $\text{NH}_4\text{-N}$  in respective order. When all the surface soils had been considered, the predicting equation relating fixed  $\text{NH}_4\text{-N}$  to silt percentage accounted for only 48% variation of fixed  $\text{NH}_4\text{-N}$ . Similarly the predicting equation relating the fixed  $\text{NH}_4\text{-N}$  percentage of silt + clay together, when all the samples irrespective of soils had been considered, was found to control only 44% variation of the total fixed  $\text{NH}_4\text{-N}$ .

### References

- Barshad, I. (1954): Cation exchange in Micaceous minerals: II. Replaceability of ammonium and potassium from vermiculite, biotite, and montmorillonite. *Soil Sci.* **78**, 57-76.
- Black, C. A. (1965): Methods of soil analysis, *Am. Soc. Agron.*, Madison, U. S. A., 1367 pp.
- Bremner, J. M., Nelson, D. A., Silva, J. A. (1967): Comparison and evolution of methods of determining fixed ammonium in soils. *Soil Sci. Soc. Am. Proc.* **31**, 462-472.
- Cottenie, A. (1980): Soil and plant testing as a basis of fertilizer recommendations. *FAO Soils Bulletin* **38** (2), Rome., 100 pp.
- Dalal, R. C. (1977): Fixed ammonium and carbon-nitrogen ratios of some Trinidad soils. *Soil Sci.* **124**, 323-327.
- Dhriwal, A. P., Stevenson, F. J. (1958): Determination of fixed ammonium in soils. *Soil Sci.* **86**, 323-327.
- Freney, J. R. (1964): An evaluation of naturally occurring fixed ammonium in soils. *J. Agric. Sci.* **63**, 297-303.
- Jackson, M. L. (1973): *Soil chemical analysis*. Prentice Hall of India (P) Ltd., New Delhi.
- Martin, A. E., Gilkes, R. J., Skjemstad, J. O. (1970): Fixed ammonium in soils developed on some Queensland phyllite and its relation to weathering. *Aust. J. Soil Res.* **8**, 71-80.
- Mikama, D. T., Kanehiro, Y. (1968): Native fixed ammonium in Hawaiian soils. *Soil Sci. Soc. Am. Proc.* **32**, 481-485.
- Moore, A. W., Ayeke, C. A. (1965): HF-extractable ammonium nitrogen in four Nigerian soils. *Soil Sci.*, **99**, 335-338.
- Opuwaribo, E., Odu, C. T. I. (1974): Fixed ammonium in Nigerian soils 1. Selection of a method or amounts of native fixed ammonium. *J. Soil Sci.* **25**, 256-264.
- Rodrigues, G. (1954): Fixed ammonia in tropical soils. *J. Soil Sci.* **5**, 264-274.
- Sah, S. C., Pasricha, N. S. (1984): Distribution of fixed ammonium in relation to soil characteristics in some soils of Punjab. *J. Indian Soc. Soil Sci.* **32**, 39-46.

- Silva, J. A., Bremner, J. M. (1966): Determination and isotope ratio analysis of different forms of nitrogen in soils. 5. Fixed ammonium. *Soil Sci. Soc. Am. Proc.* **30**, 587-594.
- Stevenson, F. J. (1959): On the presence of fixed ammonium in rocks. *Science* **130**, 221-222.
- Stewart, B. A., Porter, L. K. (1963): Inability of the Kjeldahl method to fully measure indigenous fixed ammonium in some soils. *Soil Sci. Soc. Am. Proc.* **27**, 41-43.
- Tewari, S. N., Datta, I., Deka, A. K. (1969): HF-extractable ammonium and ammonium fixing capacity in some soils of Assam. *Ind. J. Agric. Chem.* **2**, 18-21.





## *Plant physiology and biochemistry*

---

### EFFECT OF MAGNESIUM IONS ON THE ANION UPTAKE OF PLANTS

S. A. KISS<sup>1</sup>

BORSOD CHEMICAL WORKS, KAZINCBARCIKA, HUNGARY

(Received: 3 April 1987; accepted 17 June 1987)

An examination was made of the effect of magnesium ions on anion uptake. Nitrate uptake was studied on lettuces and phosphate uptake on potatoes. It was found that magnesium treatment stimulated the uptake of both nitrate and phosphate ions. An attempt is made to explain this effect by the binding of the negative charges of the ion carriers by the magnesium ions.

**Keywords:** Anion, cation, magnesium, ion uptake

#### Introduction

The uptake of various water-soluble (hydrated) ions and their penetration of the hydrophobous lipid layer of the membrane have been studied for a long time and are still an object of intense interest for researchers. According to the carrier theory proposed by Epstein (1952), the ions are transported from one side of the membrane to the other by a special carrier. In the opinion of Falk and Stocking (1976) the ion carriers do not migrate, but the ions cross the membrane due to the rotatory movement of an integrant protein. This movement is brought about by changes of conformation in the protein. According to Singer (1974) the integrant proteins which reach across the membrane only make a hingeing type of movement, as do the peripheral proteins which resemble them. The assumption is that two integrant proteins are so close to each other that a hydrophilic ion channel is created between the two surfaces. The two membrane proteins transport the ions along this ion channel to the other (inner) side of the membrane by means of hingeing movements. In either theory, energy is required for the transport of the ions (the movement of the proteins). It should be noted that there is still no unanimous opinion with respect to the ion carriers.

The energy required by the ion carriers is provided by ATP. Due to the action of ATP-ase, protons are liberated from the ATP molecules, and these migrate away from the cytoplasm (proton pump). Consequently, a difference

<sup>1</sup>Reprint request: Sándor A. Kiss, H-5900 Orosháza, Kelet u. 40. Hungary.



in potential is created between the two surfaces of the membrane; measurements indicate that this varies between 60 and 160 mV. This protein gradient enables ions to be absorbed. By means of ion exchange, cations are taken up with the help of ion carriers, in place of hydrogen ions, and anions in place of the hydroxyl ions formed as a secondary process.

Kolosov (1948) found that cations had a pronounced effect (proportionate to their concentration and charge) on the uptake of anions by the roots, as was shown by anionic (e.g. indigo carmine) pigment uptake. This has been confirmed by recent anion pigment uptake examinations (Kiss, 1986). In the light of the above, the effect of magnesium ions on the uptake of nitrate and phosphate ions was studied.

### Materials and methods

The nitrate uptake analyses were carried out on lettuce plants grown in pots in a perlite culture and watered with modified Prjanishnikov nutrient solution (Table 1). The treatments consisted of increases in the magnesium content of the nutrient solution (from 0.085 g/l to 0.17 g/l) or spraying the leaves three times with a 1% solution of  $\text{MgSO}_4$ . The nitrogen content of the plants was determined using the Kjeldahl method. The magnesium content was measured using an atomic absorption spectrophotometer. The experiments were carried out in six replications.

The phosphate uptake was studied in the field on potato plants grown in small plots, using control and  $\text{MgSO}_4$  (22 kg Mg/ha) treatments. The initial soil characteristics are shown in Fig. 2. The phosphorus and potassium contents of the soil were determined after dissolution

Table 1

*Composition of the modified Prjanishnikov solution g/l*

$\text{NH}_4\text{NO}_3$	0.240
$\text{CaHPO}_4 \cdot 2 \text{H}_2\text{O}$	0.172
$\text{CaCl}_2$	0.280
$\text{MgSO}_4 \cdot 7 \text{H}_2\text{O}$	0.085
KCl	0.360
$\text{FeCl}_3 \cdot 6 \text{H}_2\text{O}$	0.025
Hoagland's A-Z solution	1 ml

Table 2

*Soil analysis data for the potato experiment*

$\text{pH}_{\text{KCl}}$	5.93
Viscosity $\text{K}_A$	45
$\text{CaCO}_3$ %	0
Humus %	2.61
$\text{P}_2\text{O}_5$ ppm	626.0
$\text{NO}_3 + \text{NO}_2$ ppm	13.0
$\text{K}_2\text{O}$ ppm	361.0
Mg ppm	388.0

in AL-solution, while the other ions were dissolved out in 1 M KCl solution. The determinations were carried out according to specifications given by the Plant Protection and Agrochemistry Centre of the Ministry of Agriculture and Food. The phosphorus content of the potato tubers was determined after perchloric acid digestion on the basis of their molybdenum blue colour reaction using a Spekol 1 photometer. The magnesium was again measured, after preliminary reduction to ashed and hydrochloric acid uptake, using an atomic absorption spectrophotometer.

### Results

The effect of magnesium treatment on the size and mass of lettuce and lettuce leaves are demonstrated in Table 3 and Figs 1, 2. Magnesium increased both the size and the mass of the lettuce leaves.

The nitrogen and magnesium contents of the lettuce plants as a function of the treatments are shown in Table 4. The data indicate that magnesium increased the magnesium and nitrogen contents of the lettuce regardless of whether it was applied through the leaves or roots.

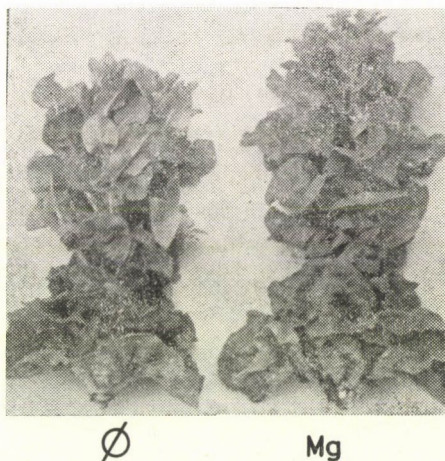


Fig. 1. Effect of magnesium treatment on lettuce

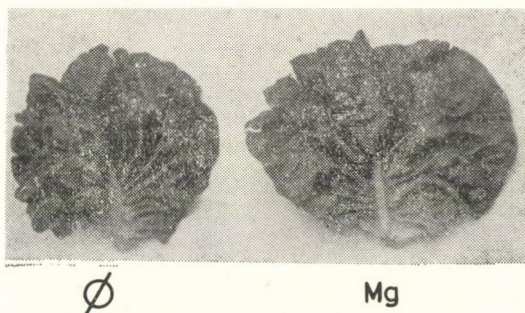


Fig. 1. Effect of magnesium treatment on lettuce leaves



Table 3

*Effect of magnesium treatment on lettuce plants*

Parameter studied	Treatment		Greatest deviation from mean	%
	0	Mg		
Leaf length, mm	116.2	121.5	3	4.6
Leaf width, mm	151.4	165.5	5	9.3
Leaf thickness, $\mu\text{m}$	349.0	331.0	6	-5.2
Height of seeded plant, cm	31.8	37.0	2	16.4
Mass of seeded plant, g	185.1	240.3	10	29.8

Note: The leaf samples were taken from identical leaf levels.

Table 4

*Effect of magnesium treatment on the nitrate and magnesium uptake of lettuce plants in terms of dry matter*

Treatment	N	Ratio	Mg	Ratio
	%			
Control	3.23	100	0.230	100
Mg through the roots	4.12	128	0.260	113
Mg through the leaves	4.28	132	0.282	123
Greatest deviation from the mean	0.19		0.011	

Table 5

*Effect of nitrogen and magnesium treatments on the tuber yield of potatoes, kg/plot*

Treatment	0 kg N/ha (control)	100 kg N/ha	Ratio, %
Control	30.25	—	100
N	—	32.0	106
N+Mg	—	36.9	122
Greatest deviation from mean		2.1	

Table 6

*Effect of magnesium treatment on the phosphate and magnesium contents of potato tubers in terms of dry matter*

Treatment	P <sub>2</sub> O <sub>5</sub>	Ratio	Mg	Ratio
	%			
Control (N)	0.421	100	0.184	100
N+Mg	0.516	142	0.223	121
Greatest deviation from mean	0.024		0.012	



The yield results for potatoes are summarized in Table 5. The magnesium treatment had a favourable effect on the quantity of yield. The phosphate uptake and magnesium content of the potato plants (tubers) are illustrated in Table 6. Examinations on potatoes indicate that magnesium treatment increased the phosphorus uptake and magnesium content of the tubers.

### Discussion

The experimental results confirmed the stimulatory effect of magnesium on the nitrate and phosphate uptake of lettuce and potato plants. Similar results were obtained by Litinski (1973) on oats, where the control plants contained 0.0353%  $P_2O_5$  compared to 0.046%  $P_2O_5$  in plants treated with magnesium ( $\Delta\% = 30.1$ ). This stimulatory effect observed in various plant species and for two types of anion uptake indicates that the stimulatory effect of magnesium on the uptake of anions can probably be generalized.

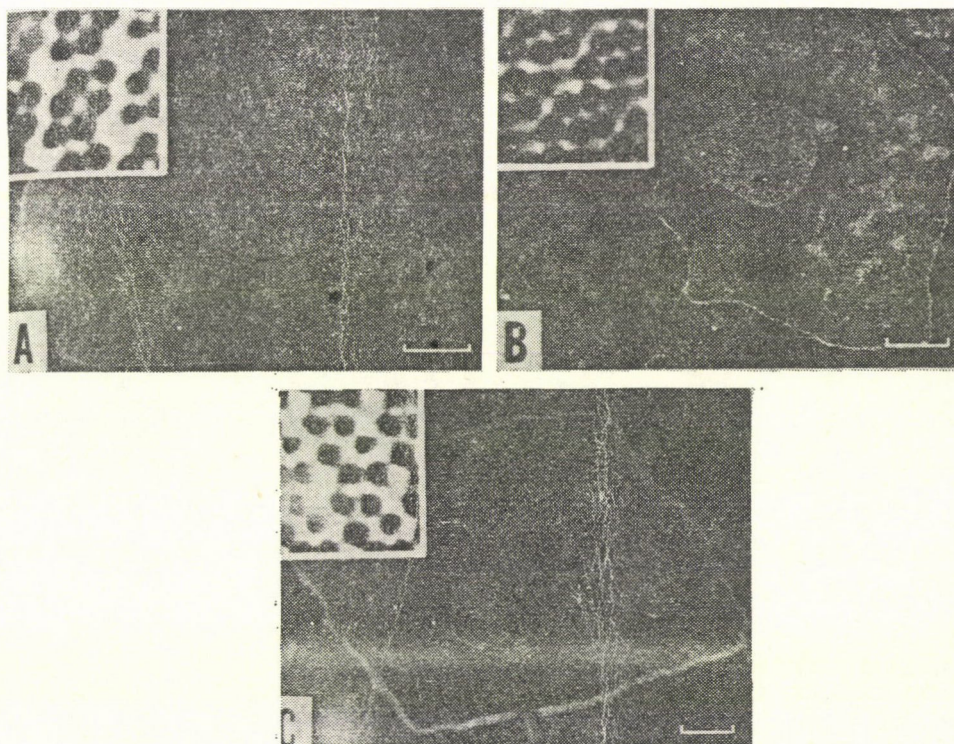


Fig. 2. Electron microscopic picture of the outer membrane of *Neurospora crassa* mitochondria (Mannella, Frank, 1982)

A: Control; B: Treated with 1mM  $MgCl_2$ ; C: Treated with EDTA



The mechanism by which magnesium promotes the uptake of anions is explained after Singer (1974) by the presence of ion channels between the membrane proteins and by the hinge-like movement and charge of these proteins.

According to Colombini (1980) and Roos et al. (1982), the ion permeability of the membrane may be regulated by the size and distribution of charges in and around the entrances of the ion channels. This possibility was studied by Mannella and Frank (1982) by means of an electron microscope (Fig. 2.). Their examinations showed that when the membranes of *Neurospora crassa* mitochondria were negatively stained with phospho-tungstic acids, the anionic dye became fixed on the basic amino acids of the proteins. In Fig. 2A six hexagonally arranged dye-fixing sties form a unit cell. Each of the dye-fixing sties marks an ion channel in the membrane. The accumulation of anionic dye molecules around the pores can be increased by the addition of magnesium (or other two-valency cations), e.g. 1 mM  $MgCl_2$  (Fig. 2B), or reduced by preliminary treatment of the membranes with EDTA (Fig. 2C). this demonstrates that the fixation of magnesium also plays a role in the charge building up in the area around the channels.

Johnston et al. (1979) and Moller et al. (1982) found that the oxidation of NADH was stimulated in intact plants and mushroom mitochondria by multivalent cations. This is explained by Johnston et al. (1979) by the fact that anionic NADH is better able to reach the dehydrogenase on the inner membrane. The possibility that cations increase the accessibility of the substrate shows that the permeability of the external mitochondrial membrane to anions has also increased, since the negative charges in or around the channels have been neutralized (Mannella, 1985).

On the basis of the above, the stimulatory effect of magnesium on the uptake of anions is explained by the hinged ion channel model set up by Singer (1974) and by the observation made by Mannella and Frank (1982) that an increase in the supply of magnesium increases the binding of negative charges in and around the ion channels. This change in the charges promotes the transportation of nitrate and phosphate ions (and of anions in general) across the membrane, i.e. it increases the anion permeability of the membrane.

It should be noted that the environment, for instance the temperature, also has a significant effect on ion uptake (Zsoldos, 1981), indicating the heat sensitivity of the membranes.

## References

- Colombini, M. (1980): Pore size and properties of channels from mitochondria isolated from *Neurospora crassa*. *J. Membrane Biol.*, **53**, 79-84.  
 Falk, H., Stocking, C. R. (1976): *Plant membranes*. *Encycl. Plant Phys.* 3, 3-50. Springer Verlag, Berlin.

- Johnston, S. P., Moller, I. M., Palmer, J. M. (1979): The stimulation of exogenous NADH oxidation in Jerusalem artichoke mitochondria by screening of charges on the membranes. *FEBS Lett.*, **108**, 28-32.
- Kiss, A. S. (1986): *A talaj savanyosodása fokozza az alumínium toxikusságát, amit a magnézium ellensúlyoz.* (Acidification of the soil increases the toxicity of aluminium, which is counteracted by magnesium.) XIV. Mezőgazdaság Kemizálása Szimpózium, Keszthely I, 11-16.
- Kolosov, I. I. (1948): Vliyanie kationov na pogloshchenie kislykh krasok i anionov mineralnykh solei kornyamin rastenii. *Trudy Inst. Fiziol. Rast. A. N. S. S. S. R.*, **6**, 1-8.
- Litinski, T. (1973): Zest' Probl. *Postepow NAUK*, **149** (23), 33-40.
- Mannella, C. A. (1985): *The outer membrane of plant mitochondria.* In: Douce, R. and Day, D. A.: Higher Plant Cell Respiration Vol. 18, 118-133. Springer-Verlag, Berlin.
- Mannella, C. A., Frank, J. P. (1982): Electron microscopic images of crystalline outer membranes from *Neurospora crassa* mitochondria. In: Le Poole, J. B.: Electron microscopy. Frankfurt.
- Moller, I. M., Schwitzgebel, J. P., Palmer, J. M. (1982): Binding and screening by cations and effect on exogenous NAD(P)H oxidation in *Neurospora crassa* mitochondria. *Eur. J. Biochem.* **123**, 204-214.
- Roos, N., Benz, R., Brdiczka, D. (1982): Identification and characterization of the pore-forming protein in the outer membrane of rat liver mitochondria. *Biochim. Biophys. Acta*, **686**, 204-214.
- Singer, S. J. (1974): The molecular organization of membranes. *Ann. Rev. Biochem.* **43**, 805-833.
- Zsoldos, F. (1981): Gyökér iontranszport változások környezeti stresszhatásokra. (Changes in root ion transport due to environmental stress effects.) *MTA Biol. Oszt. Közl.*, **24**, 227-238.





## STUDY OF FLOWERING IN PEPPER (*CAPSICUM ANNUUM* L.) GROWN UNDER CONTROLLED ENVIRONMENTS (PHYTOTRON)

Á. MÁTHÉ and K. EL-BAHADLI

UNIVERSITY OF HORTICULTURE, BUDAPEST, HUNGARY

(Received: 19 February 1987; accepted 17 July 1987)

Experiments carried out in the phytotron with sweet pepper (*Capsicum annuum* L.) have proved that the index of flowering (index-V) is suitable for following the course of flowering in this species. It was established on the basis of index-V values that the varieties "Hatvani hajtató", "Soroksári hajtató" and "California wonder" responded differently both to day-length (8, 12, 16, 24 hours) and to light intensity (5000 lux and 20 000 klux).

**Keywords:** pepper, *Capsicum annuum* L., flowering, index of flowering, "cv.", "Hatvani hajtató", "Soroksári hajtató", "California wonder"

### Introduction

In earlier papers (Máthé, 1977, Máthé and Máthé, 1979) a phenological study on wild growing (*Adonis vernalis* L.) populations was reported. Attention was called to the significant role of the "human factor" in the exact determination of the phenophases.

In order to decrease the role of subjectivity, the index of flowering (index-V) has been elaborated rendering it possible for the phenophases of flowering, budding and fruit setting to be numerically determined. Its value, as verified in further studies on *Matricaria chamomilla* L. (Franz et al., 1985), might also be correlated with the essential oil content of the plant, thus giving it practical implications. The index provides values for the more exact demarcation of the phenophases, thus facilitating a more reliable comparison of treatments. The numerical expression of the phenophases can also play an important in experiments conducted in the phytotron. Therefore, in ecological investigations on sweet pepper (*Capsicum annuum* L.) studies were made on the applicability of the index-V, as an experimental tool in evaluating experiments on the initial stages of flowering.

### Materials and methods

In the experiments, the following three sweet pepper varieties of different growth type were used: "Hatvani hajtató", "Soroksári hajtató" and "California wonder".

The experiments were carried out under controlled environmental conditions in the phytotron (type: Conviron G-30, made in Canada) of the Institute of Vegetable Cultivation,



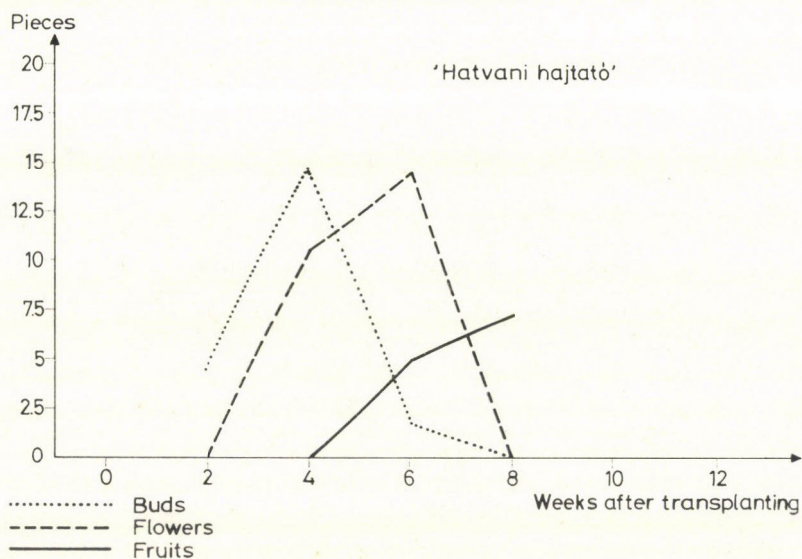


Fig. 1. Flowering curve of *Capsicum annuum* L. cv. "Hatvani hajtató"

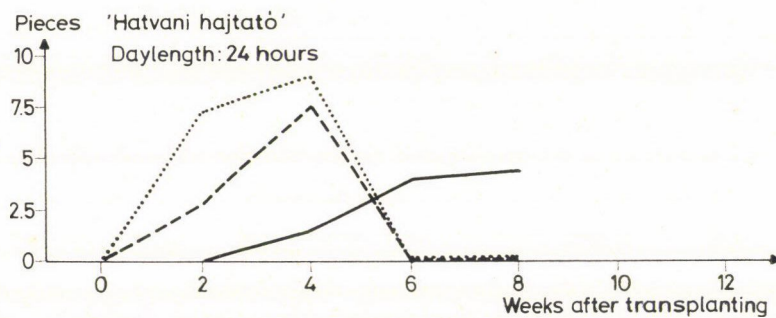
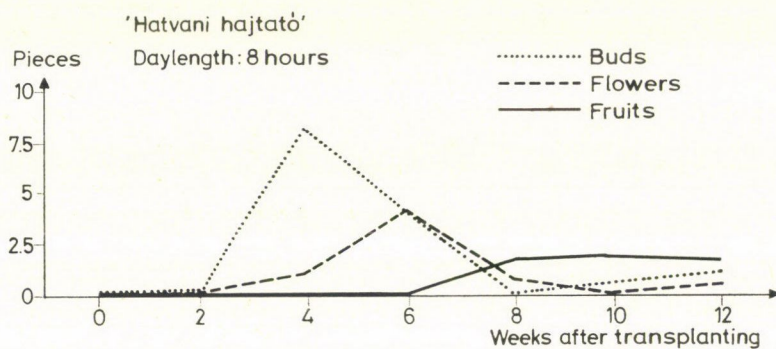


Fig. 2. The effect of daylength on the flowering curve of *Capsicum annuum* cv. "Hatvani hajtató"

University of Horticulture, Budapest. The plants were propagated generatively by raising transplants. Seeds were sown into multiplying boxes filled with a 2 : 1 mixture of peat and sand. From the time of seedling emergence a commercially available 0.5% Volldünger solution was applied. The temperature was kept at 30 °C during germination and 25 °C during emergence. During the day the light intensity was kept at 10 Klux.

After reaching the 4-leaf stage the transplants, in 10 × 10 cm plastic boxes, were transferred to the phytotron, where the following treatments were applied: temperature 20 °C (constant); light was provided by Sylvania CW type incandescent tubes and TUNGSRAM 100 W lamps; light intensity was 5 Klux and 20 Klux; 70%.

In order to characterize flowering, the number of buds (*b*), flowers (*v*) and fruit with a size of 2 cm (*t*) was recorded at weekly intervals. The mean of 6 plants was tabulated according to the index of flowering (*V*):

$$\text{index-}V = \frac{t - b}{b + v + t}$$

The value of index-*V* varies between -1 and +1, expressing the phenophases of budding and fruit-set, respectively. The value 0 expresses the phenophase of full flowering, when the number of flowers is the highest.

### Results and discussion

The aim of the experiments was to make observations on the flowering characteristics of sweet pepper grown under controlled environmental conditions. To this end, as a first step, the course of development of the generative organs (buds, flowers, fruit) of the variety "Hatvani hajtató" was studied in the 20 Klux, 16-hour day-length treatment at weekly intervals. As seen in

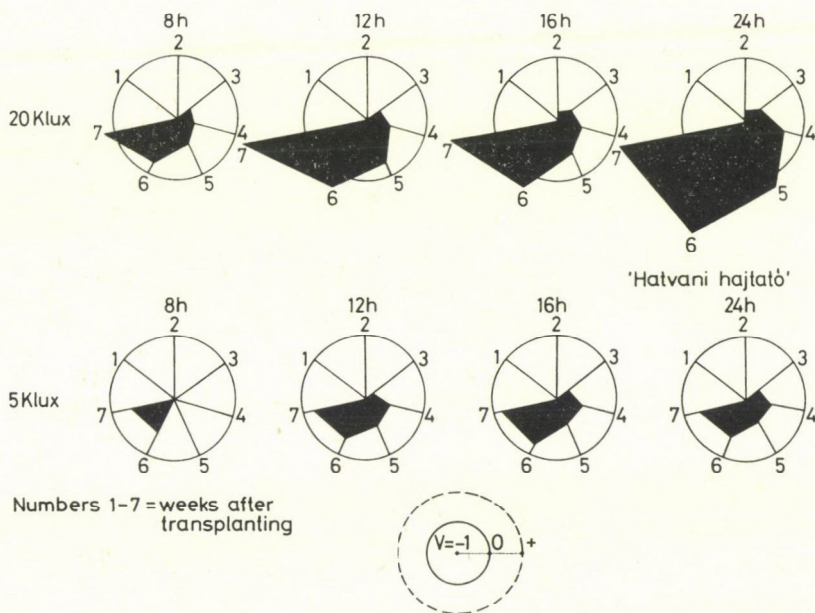


Fig. 3. The course of flowering of *Capsicum annuum* L. cv. "Hatvani hajtató" as affected by daylength and light intensity.



Figure 1, the increasing number of buds peaks in the 4th week. Flowering starts in the 2nd week, reaching a maximum in the 6th week, at a date when the decreasing frequency of buds and the increasing frequency of fruit is at equilibrium.

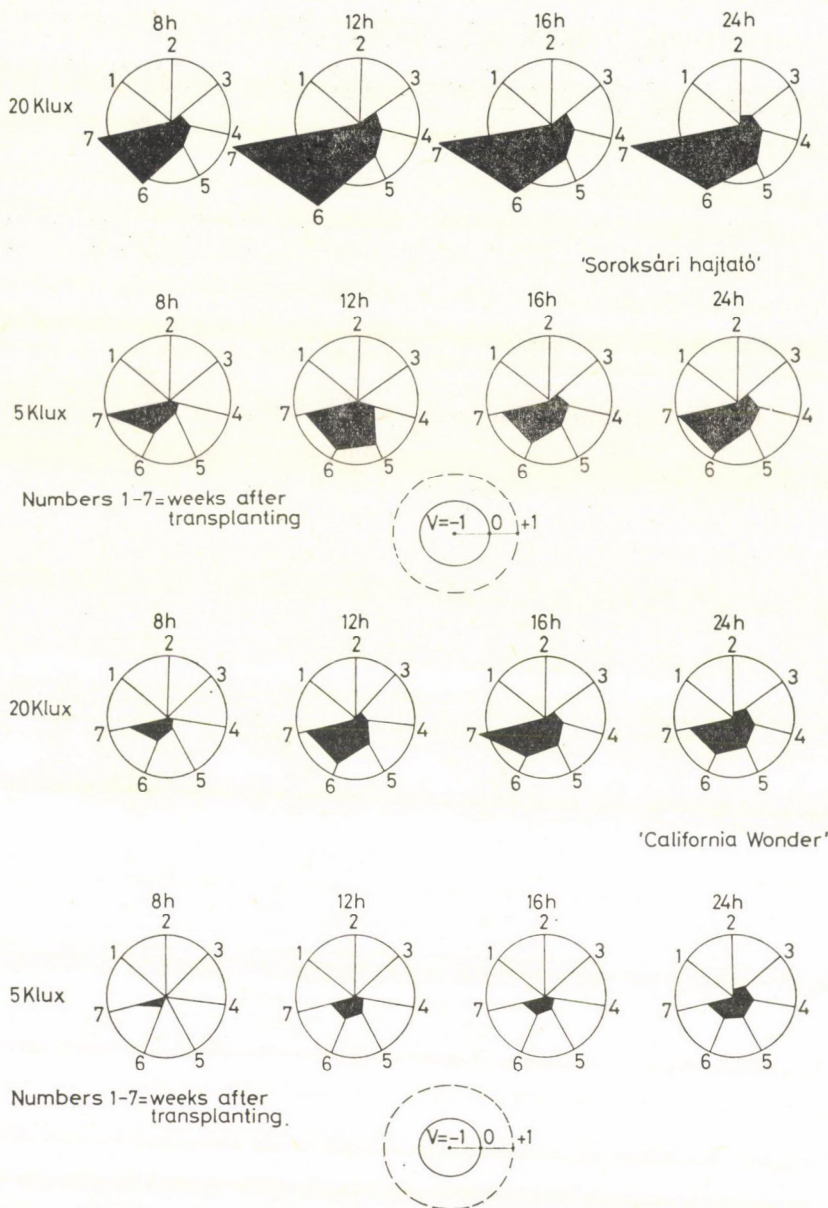


Fig. 4. The effect of light intensity and day-length on the flowering of *Capsicum annuum* L. cv. a. "Soroksári hajtató" b. "California wonder".

The above characteristics of flowering are similar to those observed in the case of *Adonis vernalis* L. (Máthé, 1977, Máthé and Máthé jr. 1979), indicating that index-V could be suitable for describing the course of flowering in *Capsicum annuum* L., too.

In Figure 2, it can be seen that under unfavourable ecological conditions (8- and 24-hour day-length) the variety "Hatvani hajtató" does not display the characteristic or "ideal" pattern of flowering observed in the 16-hour treatment. According to this latter pattern, the decreasing frequency of buds crosses the ascending curve of fruit frequencies at a point (date) when the frequency of flowers culminates. At this phase the plants are in full bloom and the value of index-V equals 0.

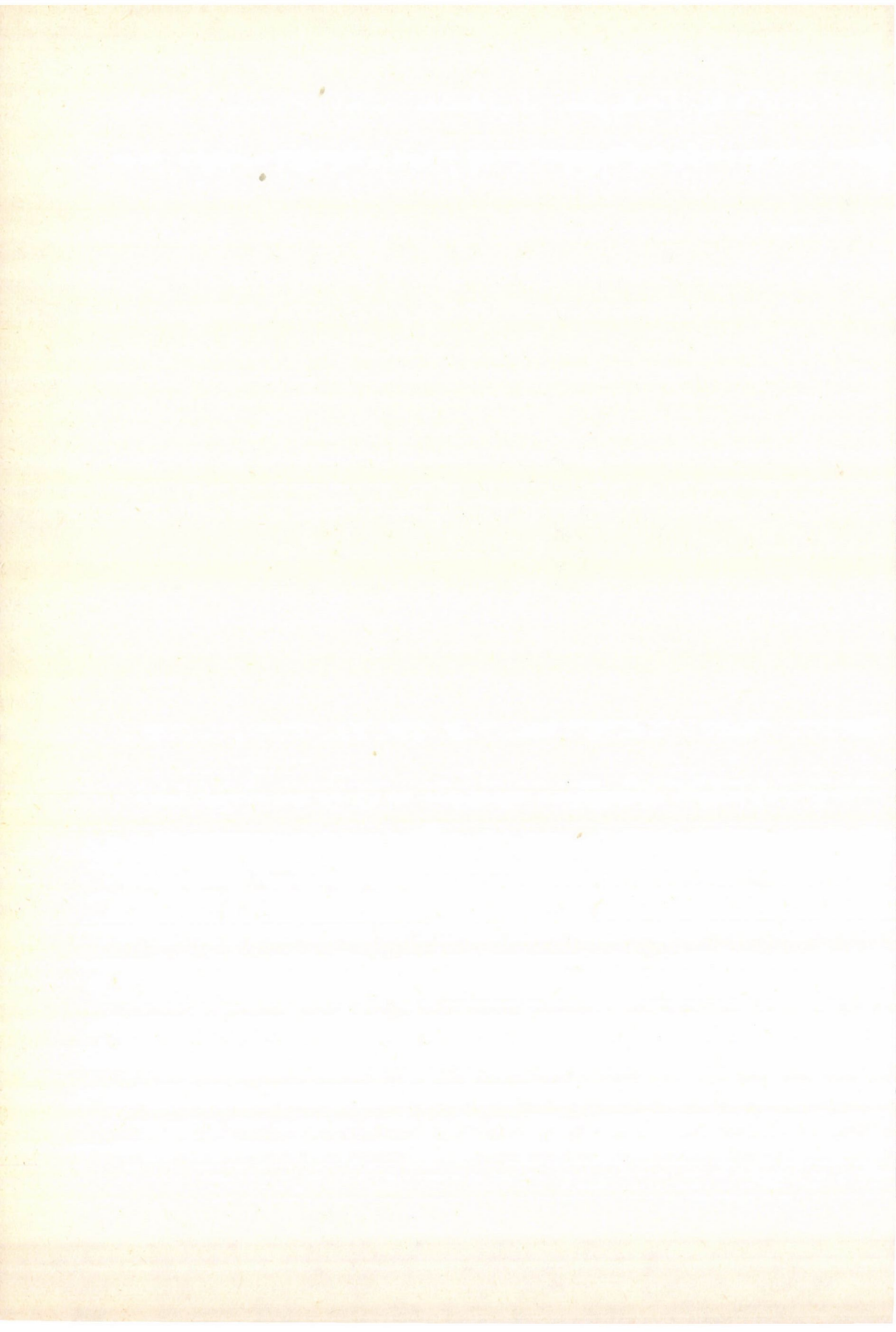
In Figure 3, the entire experiment is characterized by means of the flowering index. The circle diagrams facilitate an overall view of the treatments. It can be seen that a light intensity of 20 Klux is much more favourable for the variety "Hatvani hajtató" than 5 Klux. In the latter treatment, the plants did not reach the phenophase of full flowering at all, while under 20 Klux illumination, full flowering varied between the 5th to 7th weeks after transplanting.

The differences are even more explicit, when the varieties "Soroksári hajtató" and "California wonder", which have different growth types, are compared (Figure 4), since the latter only reached full flowering at a 16 hour day-length in the 20 Klux treatment. The variety "Soroksári hajtató" responded very sensitively to the 5 klux treatment. This is verified by the fact that the index-V did not reach a value of 0, i.e. full flowering. In contrast to this, under the 20 Klux conditions, full flowering could be observed between the 6th to 7th weeks, in all the day-length variations.

### References

- Franz, Ch., Hölzl, J., Máthé, Á., Winklhofer, A. (1985): Recent results on cultivation; harvest time and breeding camomile. *Chamomile in Industrial and Pharmaceutical Use*. Triest. 6-17.
- Máthé, Á. (1977): Az *Adonis vernalis* L. virágzásának számszerű kifejezése. (Numerical expression of the flowering of *Adonis vernalis* L.). *Herba Hung.* 16, 35-47.
- Máthé, Á., Máthé, I. jr. (1979): Study of the flowering and generativity of *Adonis vernalis* L. populations. *Acta Bot. Hung.*, 25, 83-87.





## WAYS OF PREDICTING STORAGE LOSSES IN APPLES

P. SASS and Z. LAKNER

UNIVERSITY OF HORTICULTURE AND FOOD INDUSTRY, BUDAPEST, HUNGARY

(Received: 1 April 1987; accepted 30 July 1987)

A prediction of the storage losses for Jonathan apples would be very important from a practical viewpoint. This paper explores the more profound correlations between the losses arising in the course of storage and the physical and chemical parameters of the fruit at the beginning of storage. Multivariable stepwise analysis and path analysis were used in the calculations. The numerical relations deduced from the correlations between the factorial systems, also illustrated graphically, could make a significant contribution to the more accurate prediction of storage losses.

**Keywords:** Apple storage, systems approach, Jonathan apples, Jonathan spottedness, mass loss, rotting, internal breakdown, quality parameters at beginning of storage

### Introduction

The losses arising in the course of long-term storage can be divided by and large into three major groups: (a) mass loss, (b) diseases induced by physiological changes, and (c) rotting caused by fungal diseases. The causes or components of each source of loss are extremely complex. Some can be attributed to environmental and production factors, while others result from changes occurring during storage.

Thus, refrigerated storage is a complex process, which can be described using the concepts and conclusions associated with the general theory of systems (Csáki, 1971; Jándy, 1981).

In the light of this, from the point of view of system control, the problem with respect to the refrigerated storage model examined can be interpreted by taking the values of the  $x(t)$  control variable as completely identical with the values of the  $n(t)$  input vector the aim is to find values of the  $n(t)$  vector where the components of the  $q(t)$  vector, related to the parameters of products taken out of storage and determined by the function set  $z(t)$  describing the trend in storage losses as a controlled variable, have optimum values. In other words, if the parameters characteristic of the quality expected of products taken out of storage are determined and the storage conditions are accurately recorded (temperature, humidity, refrigeration rate, atmospheric composition and the constancy of these parameters), the efficiency of the storage process is deter-



mined by the quality of the products put into storage. This statement is equivalent to saying that under fixed storage conditions the success of storage depends exclusively on the date when the products are taken out of storage and on the quality of the products originally stored.

Due to the known characteristics of apples, only certain properties can be determined and described when characterizing the quality of the fruit put into storage. The following requirements can be raised with respect to these properties:

- they should give as clear as possible a description (as much information as possible) of the apples to be stored;
- the measurement of the chosen parameters should be relatively simple (the application of numerous complex analytical methods must thus be dispensed with);
- the values of the parameters determined should be objective, i.e. they should depend as little as possible on the person carrying out the analysis.

By applying the apple quality indices obtained using the above criteria it is possible to calculate a correlation between the quality indices of the apples at the beginning and end of storage, and of the storage losses for apples taken out of storage.

The practical applicability of calculations carried out in this way is determined by three factors:

- the closeness of the correlation between the quality parameters measured on apples put into storage and the losses arising during storage, i.e. the reliability of the prediction;
- the number of parameters required for the prediction of the expected losses with the desired reliability (the fewer parameters it is necessary to know or determine, the greater the chance that the classification method elaborated will be put into practice).

### Materials and methods

Research has been in progress at the Fruit Production Department of the University of Horticulture and Food Industry for the last twenty years on the complex analysis of the storability of the apple varieties in general cultivation and of the factors affecting the success of storage.

The physical and chemical parameters and the healthness of apples at the beginning and end of storage were characterized with the aid of a total of 55 parameters, and efforts were made to ensure that the apple lots tested were a truly representative sample. Thus, over the years, several million data have accumulated for each variety. The logical scheme of the examinations is shown in Fig. 1.

The mass of data acquired during the experiments was analysed first with single factor and later with multifactorial regression analyses. The results of these calculations have been reported in a number of publications (Sass 1982, 1986).

The major functions from the standpoint of prediction are as follows:

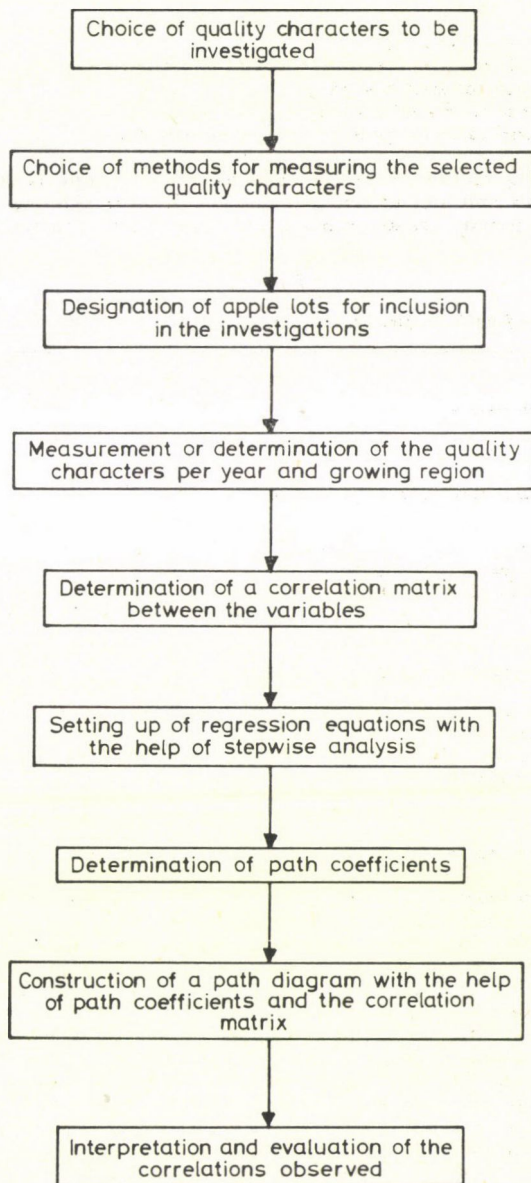


Fig. 1. Block diagram illustrating the course of the examinations

$$\begin{aligned}
 Y_{31J} &= 10.515 - 0.024 Y_{16} - 0.173 Y_{46} \\
 Y_{1J} &= 12.133 + 0.111 Y_{16} - 0.012 Y_{44} - 0.822 Y_{46} \\
 Y_{5J} &= -6.194 + 0.070 Y_{23} \\
 Y_{9J} &= -21.036 + 0.192 Y_{23}
 \end{aligned}$$

where:

$Y_{31J}$  = mass loss of Jonathan apples  
 $Y_{1J}$  = Jonathan spottedness



$Y_{5J}$  = Jonathan rotting  
 $Y_{9J}$  = Jonathan breakdown  
 $Y_{16}$  = mean colouring of the apples when put into storage  
 $Y_{23}$  = mean mass of apples when put into storage  
 $Y_{44}$  = acid content of apples when put into storage  
 $Y_{46}$  = sugar/acid ratio of apples when put into storage

In the present paper, these correlation analyses are further developed and the connection between the dependent and independent variables of the regression equations describing the relation between the parameters determined for apples at the beginning of storage and the

**Table 1**  
*Role of path coefficients in loss predictions for Jonathan apples*

Regression relation	Formula	Absolute correlation value	Relative weight no. % as a % of total variance
Direct relation between mean colouring and mass loss	$P_1$	= 0.061	6.1
Indirect relation between mean colouring and mass loss through factor $Y_{46}$	$P_2 \cdot r_{12}$	= 0.055	5.5
Direct relation between sugar-acid ratio and mass loss	$P_2$	= 0.084	8.4
Indirect relation between sugar-acid ratio and mass loss through factor $Y_{16}$	$P_1 \cdot r_{12}$	= 0.055	5.5
That part of the variance of the equation describing the relation between mass loss, sugaracid ratio and mean colouring which can be explained by the two factors above	$r^2$	= 0.2	20

losses or diseases arising in the course of storage are analysed using the path analysis method. This biometric method, which is only applied relatively rarely at present, makes it possible to devise an accurate description of the interactions and relationships existing between the variables of the regression functions.

The role played by the path coefficients in loss predictions is illustrated for the regression equation describing the relationship between the mass loss in Jonathan apples ( $Y_{31J}$ ), the sugar-acid ratio when put into storage ( $Y_{46}$ ) and the mean colouring ( $Y_{16}$ ) (Table 1).

The calculated interactions and correlations are illustrated in the path diagram in Fig. 2. This is fundamentally a directed graph. The closeness of the interactions is also indicated by the thickness of the various lines in the graph, which is proportional to the relative path coefficient.

The path diagram shown in Fig. 3. was also compiled with the help of path analysis. This diagram illustrates the interaction between the quality parameters measured at the beginning of storage and the relationship between parameters measured prior to storage and the storage losses found at the end of storage, based on the regression equations determined.

This type of breakdown means that improvements in the efficiency of storage techniques, i.e. smaller losses, can be achieved by deliberate efforts to alter certain parameters measured at the beginning of storage.

## Discussion and conclusions

The rotting caused by fungi and internal breakdown are closely correlated with each other. Both factors are significantly correlated with the mean mass of the apples put into storage. The development of fungal diseases and

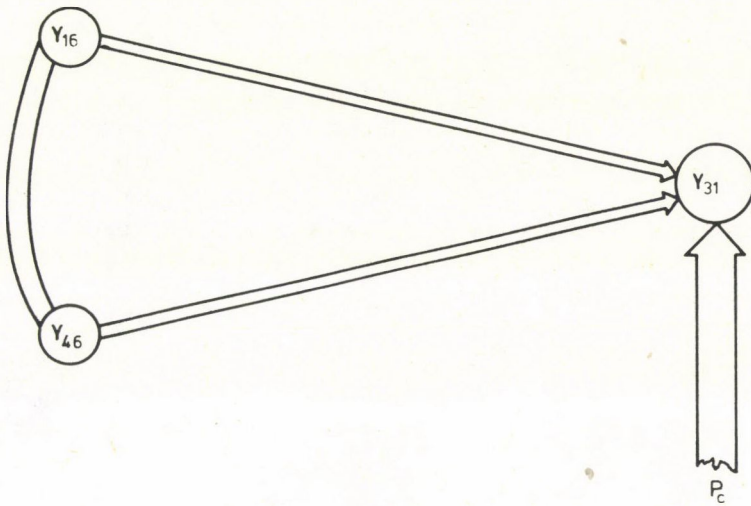


Fig. 2. Path diagram of factors influencing the mass loss arising during the storage of Jonathan apples  
 $Y_{16}$  mean colouring,  $Y_{46}$  sugar-acid ratio at beginning of storage;  $Y_{31}$  mass loss;  $P_c$  effect of other factors

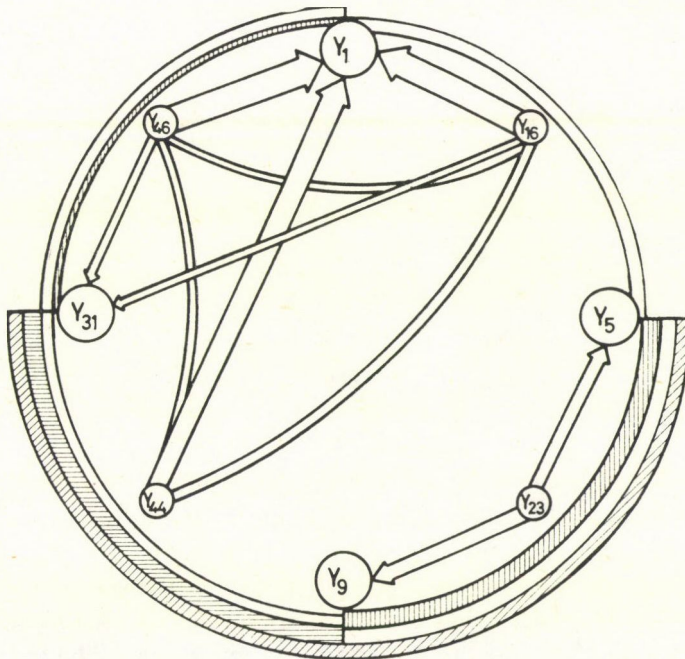


Fig. 3. Factors influencing storage losses in Jonathan apples  
 $Y_1$  Jonathan spottedness;  $Y_5$  rotting;  $Y_9$  internal breakdown;  $Y_{31}$  mass loss;  $Y_{16}$  mean colouring;  $Y_{23}$  mean mass;  $Y_{44}$  acid content at beginning of storage;  $Y_{46}$  sugar-acid ratio at beginning of storage



internal breakdown is influenced by many factors simultaneously. It is difficult to predict storage losses arising in this way, but a knowledge of the mean mass provides a certain basis for such predictions. There is a less close relation between mass losses arising during storage and the occurrence of Jonathan spottedness. It is noteworthy that in the regression equations the mean colouring and the sugar-acid ratio at the beginning of storage are found among the explanatory factors for both sources of loss. These parameters can be applied more accurately for the prediction of Jonathan spottedness than for that of mass loss.

There is an extremely close correlation between the sugar-acid ratio and the sugar content, so in the regression equations the sugar-acid ratio can be regarded as approximately equivalent to the sugar content.

It can be stated that the sugar-acid ratio and the acid content together provide an interpretation of a substantial part of the known (explained) variance of the Jonathan spottedness arising. Both factors have negative path coefficient values, which means that the direct, and to some extent the indirect, effect of these factors reduces the variance of Jonathan spottedness; while mean colouring, which has a positive path coefficient, increases the variance of the Jonathan spottedness arising. The sugar-acid ratio is also significantly correlated with the mass loss observed. Due to the negative path coefficients, both explanatory variables reduce the variance of mass loss.

It follows from the calculations that a determination of the sugar and acid contents is sufficient in the case of both Jonathan spottedness and mass loss to provide a partial explanation of trends in the loss factors examined. Mean colouring plays an important role (25–30%) in the prediction of these loss factors.

In summary, the following conclusions can be drawn.

It has been successfully proved that the system of concepts employed by the general theory of systems and the direction theory can be applied in examinations on the refrigerated storage of horticultural products. The use of the terms employed by cybernetics and the direction theory makes it possible to study the processes occurring in the course of refrigerated storage more accurately.

- Regressional relations were found between the losses arising in Jonathan apples during storage and certain parameters which can be simply determined at the beginning of storage. In this way it proved possible to determine which of the numerous quality characters determining the quality of apples at the end of storage were the most important;
- By using path analysis to breakdown the equations determined in the course of regression analysis, the significance of the individual quality characters measured at the beginning of storage was determined with respect to their role in the development of storage losses. These inter-

- actions were illustrated on graphs providing a more accurate and descriptive characterization than was previously possible of the relationship between apple properties measured at the beginning of storage and the storage losses observed when the apples are taken out of storage;
- The investigations carried out so far indicate that similar calculations would be justified to determine the losses arising during refrigerated storage in other varieties of apples and for other fruit.

### References

- Csáki, F. (1971): *Általános szabályozásmélet*. (General theory of regulation.) Akadémiai Kiadó, Budapest.
- Jándry, G. (1981): *Rendszerelemzés és operációkutatás*. (Systems analysis and operation research.) Műszaki Könyvkiadó, Budapest.
- Sass, P. (1982): *Az alma tárolhatóságát meghatározó törvényszerűségek a fajta, a termőtaj és a tárolási módszerek összefüggései alapján*. (General laws determining the storability of apples on the basis of correlations between the variety, the growing region and the storage methods.) Theisis, Budapest.
- Sass, P. (1986): *Gyümölcstárolás*. (Fruit storage.) Mezőgazdasági Kiadó, Budapest.





## INVESTIGATION OF THE CHEMICAL COMPOSITION-CHANGES IN HORTICULTURAL PLANTS AS A FUNCTION OF X-RAY STIMULATION DOSES

A. S. SZABÓ and M. A. J. TEJEDA

UNIVERSITY OF HORTICULTURE AND FOOD INDUSTRY, FOOD CHEMISTRY DEPARTMENT,  
BUDAPEST, HUNGARY

(Received: 9 June 1987; accepted 14 September 1987)

In the low dose experiments on radish, lettuce, tomato, paprika, pea and bean the presowing stimulation does not influence the chemical parameters (e. g. protein, fat, fibre, ash, macro and trace elements) of the edible parts of the plants. The stimulation has a favourable (positive) effect on the vitamin-C (ascorbic acid + dehydroascorbic-acid) content of the horticultural vegetables. The irradiation of the seeds was performed in the 5-15 Gy dose interval, using a mobile X-ray generator, GIGANT-18, with dose-intensity of 2 Gy/min.

**Keywords:** ascorbic acid, bean, chemical composition, dehydro ascorbic acid, low dose irradiation, radiostimulation, radish, X-ray treatment

### Introduction

In this paper we do not want to discuss the agricultural importance and applicational possibilities of radiostimulation — presowing irradiation of the seeds — because there are many publications concerning the biochemical basis of stimulation and the effect of stimulation on the yield and other parameters (Berezina, 1975; Pál, 1975; Pál and Simon, 1976; Simon 1977; Luckey, 1980; Köteles, 1982; Siciu and Draganescu, 1982). But these data are based mainly on gamma-ray treatments; and on X-ray stimulation and its effect on the chemical composition changes much less data exist.

In this article we would like to give a short account of our investigations carried out for the determination of the chemical composition-changes that depend upon the stimulation dose. We reported these results at ESNA (European Society of Nuclear Methods in Agriculture) conferences (Szabó et al. 1983, 1984, 1985).

### Materials and methods

As test plants we used radish (*Raphanus sativus*), green-paprika (*Capsicum annuum*), tomato (*Solanum Lycopersicon esculentum*), pea (*Pisum sativum*), bean (*Phaseolus vulgaris*) and lettuce (*Lactuca sativa*). The plant cultivation was carried out on small parcels with control and treated seeds.



For the presowing irradiation of the seeds — radiation treatment the day before the sowing — we used an X-ray apparatus, GIGANT-18. This mobile X-ray equipment was manufactured by the Hungarian industrial firm TRAKIS. The irradiation capacity (Dose rate) of the apparatus is 2 Gy/min. the applied doses were between 5 and 15 Gy.

The chemical analyses of the plants (grown up from the seeds) were carried out at the end of their vegetation period, on the Food Chemistry Department of the University of Horticulture, in the Center of Food Control of the Hungarian Ministry of Agriculture and Food (Budapest), Research Institute for Animal Nutrition (Herceghalom) and Central Research Institute for Food Industry (Budapest). The following measurements were performed: dry material; fat; protein; amino acid composition, fibre; N-free extract; ash; some macro and trace elements; vitamin-C.

## Results and discussion

About the chemical composition of the investigated plants (edible parts) we have much information, but of course it is impossible to include it all in this paper. Taking into consideration the natural biological deviations between the samples and the error in the analytical determinations, in most cases there were no significant differences between the measured chemical parameters of the control and the stimulated plants. For characterization of them measured differences, in Table 1 are shown some average parameters for the bean samples, cultivated in 1983.

Also the data given in Table 1 prove that the presowing radiostimulation of the seeds does not influence the most important chemical parameters of the investigated horticultural plants. The difference is significant only in the vitamin-C (ascorbic acid + dehydro ascorbic acid) content. For example to the

Table 1

*Mean chemical composition of bean samples (g/kg) in dry material*

Parameter	Control	5 Gy	10 Gy	15 Gy
Protein	280	269	276	295
Fat	16	15	16	17
Fibre	42	43	40	35
N-free extract	624	636	632	617
Ash	38	37	36	36

Table 2

*Ascorbic-acid and dehydro-ascorbic-acid content of radish*

Dose	Vitamin-C	Ascorbic-acid	Dehydro-ascorbic-acid
0	153+12	42+3	111+2
5	123+10	38+4	88+3
10	203+21	34+2	170+8
15	234+17	114+8	121+7

**Table 3**  
*Vitamin-C content of early red radish (Bálint et al., 1971)*

Dose, Gy	Vitamin-C content mg% in percent of control	
	greenhouse	field
Control	33.07	29.46
3	102.8	107.9
10	126.1	111.1
15	138.3	126.2
20	136.6	110.8
30	99.9	114.6

measurement of radish, 1983 — Table 2 — it is obvious that the result is much higher in the case of 10 and 15 Gy stimulated plants than in the control, or in those that received the 5 Gy dose. This indicates that the stimulation has a favourable effect on the vitamin-C content.

We would like to mention, that Bálint et al. (1971) published interesting data on the vitamin-C concentration changes that depend upon the stimulation dose with gamma-rays ( $^{60}\text{Co}$ ) To the results — Table 3 — from the standpoint of vitamin-C content, the optimum dose is 15 Gy for early red radish.

### References

- Bálint, A., Simon, J., Menyhért, Z., Viglási, P., Pannonhalmi, K. (1971): Radiostimulation experiments on early red-radish. *Stimulation Newsletter* 3, 41–48.
- Berezina, N. M. (1975): *Presowing irradiation of seeds of agricultural plants*. Oak Ridge National Laboratory, U. S. Atomic Energy Commission.
- Köteles, G. J. (1982): New aspects of cell membrane radiobiology and their impact on radiation protection. *ESNA Newsletter*, 4–10.
- Pál, I. (1975): Investigations on the effects of seed irradiation of plants in a phytotron. *Stimulation Newsletter*, 8, 23–36.
- Luckey, T. D. (1980): *Hormesis with ionizing radiation*. CRC Press, Boca Raton, Florida.
- Pál, I., Simon, J. (1976): Investigations on the influence of the presowing gamma irradiations ( $^{60}\text{Co}$ ) on bean plants (*Phaseolus vulgaris* L.). *Stimulation Newsletter*, 9, 46–57.
- Simon, J., Bhattachariya, S. (1977): *The present status and future prospect of radiation stimulation in crop plants*. Budapest.
- Suciu, Z., Draganescu, I. (1982): Studies concerning the influence of gamma rays on onion and carrot yields. *ESNA Newsletter*, 49–52.
- Szabó, A. S., Simon, J., Pál, I. (1983): *Investigation of the effect of X-ray stimulation on the chemical composition of some plants*. Proc. ESNA XIVth annual meeting, 5–9 Sept., Madrid, Spain.
- Szabó, A. S., Simon, J. (1984): *Chemical composition changes in dependence of radiostimulation doses by horticultural plants*. Proc. ESNA XVth annual meeting, 3–7 Sept., Piacenza, Italy.
- Szabó, A. S., Tejeda, M. A. J., Simon, J. (1985): *Investigation about the vitamin-C content changes in vegetables as a function of stimulation doses*. Proc. ESNA XVIth annual meeting, 9–13 Sept., Warsaw, Poland.





## EFFECT OF IONIZING RADIATION ON THE RESPIRATION INTENSITY OF PEARS DURING STORAGE

MAHFOUZ AL BACHIR<sup>1</sup> and P. SASS<sup>2</sup>

<sup>1</sup> SYRIAN NUCLEAR POWER AGENCY, DAMASCUS, SYRIA

<sup>2</sup> UNIVERSITY OF HORTICULTURE AND FOOD INDUSTRY, BUDAPEST, HUNGARY

(Received: 24 November 1986, accepted 17 March 1987)

According to the results of a 3-year series of experiments on ionizing radiation (<sup>60</sup>CO and X-rays) a relationship exists between the radiation doses chosen (40, 60, 100, 500, 1000, 1500 Gy) and changes in the quality of the fruits varieties. Radiation was generally found to have a stimulatory effect on the ripening processes. This is particularly so for fruits at a stage of ripening less suitable so for storage. The acceleration of ripening takes place for a short time (5-7 days) immediately after irradiation, as proved by respiration and enzyme activity tests.

When the physiological conditions during storage are taken into consideration, it can be established that on removal from storage the rate of respiration of treated fruits was lower both in controlled and in constant atmosphere, which suggests that irradiated fruits can be stored for a longer time.

**Keywords:** storage, ionizing radiation, respiration intensity, apples, pears, X-rays, gamma rays, enzyme activity, fruit ripening

### Introduction

Extending the shelf-life of fruit is a common ambition all over the world, justified by the demand for more up-to-date nutrition.

Methods and procedures whereby the shelf-life of fruit can be extended are the subject of research not only in Europe but also in other countries.

One of the preconditions of success is a knowledge of the basic life processes of fruit i.e. the metabolic processes (including respiration) which give a fair indication of the quality of the fruit at a given point of time.

In the course of studies on the biology of ripening, changes in the respiration intensity of pomiferous plants in response to ionizing radiation were traced over a period of 3 years.

The respiratory metabolism of plant tissues generally decreases under the influence of radiation though in some cases it may increase. The information found in the literature is inconsistent in this respect.

- Most authors (Meisel et al. 1954) consider an increase in the intensity of respiration to be a sign of the oxidative process taking place in the respiration substrate.
- Others think that radiation upsets the balance between oxidation and



Table 1

*Effect of various types and rates of ionizing radiation on the respiration of Jonathan apples  
(mg CO<sub>2</sub>/24 h/kg fruit)*

Type of radiation	Rate	1982			1983			1984		
		A	B	C	A	B	C	A	B	C
Control	—				659	789	848	807	855	892
X-ray	40 Gy				680	787	821	828	840	937
X-ray	60 Gy				772	800	744	842	884	856
X-ray	100 Gy				774	785	898	883	851	905
	SzD <sub>5%</sub>				42	26	47	19	23	27
Control	—	726	823	770	669	789	848	807	855	892
Gamma	500 Gy	930	773	777	1030	711	810	1099	814	798
Gamma	1000 Gy	897	737	734	1035	782	838	1108	805	846
Gamma	1500 Gy	1014	827	857	1069	784	861	1120	838	916
	LSD <sub>5%</sub>	26	27	74	31	29	24	37	26	

Note: A = at beginning of storage  
B = in the middle of storage  
C = at end of storage

Table 2

*Effect of gamma radiation on respiration in apples  
(mg CO<sub>2</sub>/24 h/kg fruit)*

Rate of radiation	At beginning	In the middle	At end
	of the storage		
Ø	734	820	837
500 Gy	1020	766	795
1000 Gy	1013	774	812
1500 Gy	1068	817	878
LSD <sub>5%</sub>	108	273	75

Note: Gy = gray (= 1 J/kg)

phosphorylation, with the result that the energetic efficiency decreases (Metlitsky and Salkova 1961).

- According to a third view (Romani 1966 in Metlitsky 1975) the more intensive respiration observed in irradiated fruit is due to the adaptation of the organism to the higher energy production required to make up for the damage that radiation has caused to the cells.

This latter view is supported by the data of experiments in which the utilization of amino acids in protein synthesis was found to increase after fruit were irradiated. It was experimentally demonstrated that after the

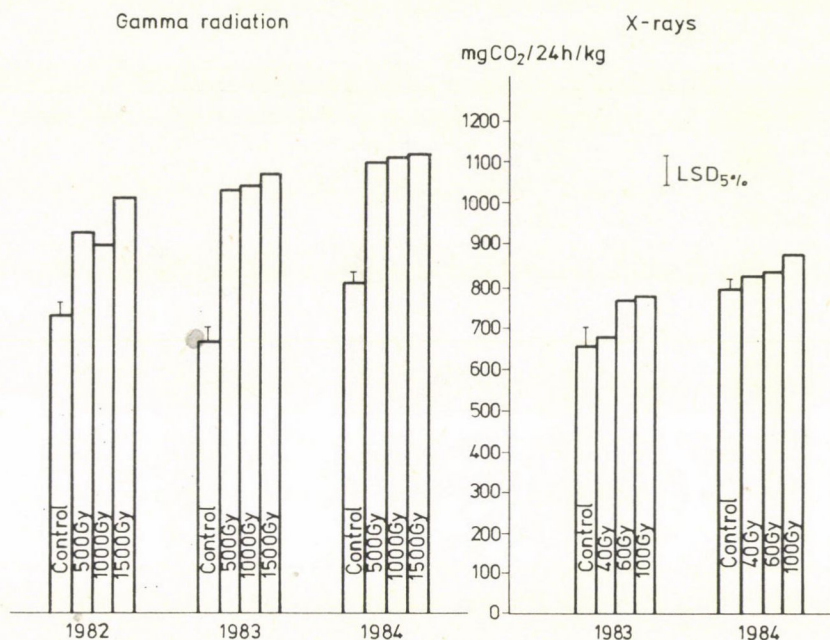


Fig. 1. Changes in the respiration of apples at the beginning of storage in response to X-rays and gamma radiation

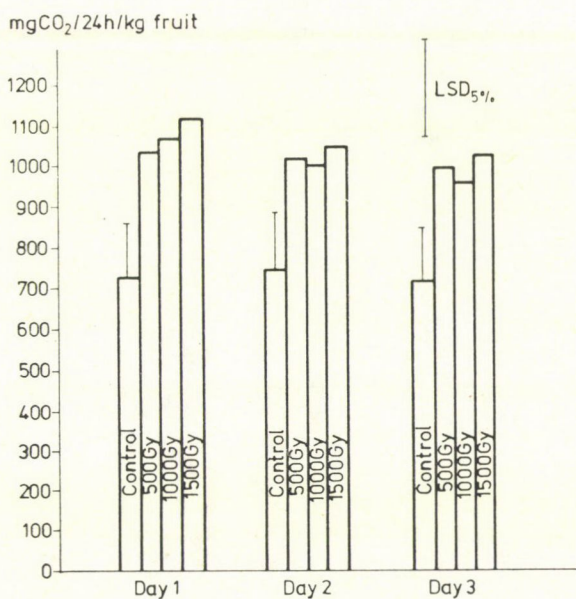


Fig. 2. Changes in the intensity of respiration of apples as a function of the rate of radiation and the length of time after the treatment (3-year average)



Table 3

*Changes in the respiration of pears at the beginning*

Variety	Treatment	1982				
		1st day	2nd day	3rd day	Average	1st day
Hardenpont	Ø	434	428	415	426	653
téli vajkörte	1000 Gy	658	648	632	646	738
Serres	Ø	372	344	308	341	441
Olivér	1000 Gy	636	641	553	610	713
Téli	Ø	394	365	299	353	433
esperes	1000 Gy	579	594	516	563	628
Nemes	Ø	371	355	318	348	426
Krasszán	1000 Gy	568	583	510	554	654
LSD <sub>5%</sub>			53			

Note = respiration CO<sub>2</sub> mg/24 h/kg fruit

radiation treatment the quantity of mitochondria in the tissues sharply decreased at first followed by an increase, reaching the value of the control after 6–7 days.

In connection with changes in the concentration of free amino acids, it should be mentioned that radiation does not influence the total protein content. This means that processes other than the decomposition of protein are responsible for changes in the amount of free amino acids. Radiation may increase the quantities of amino acids involved in oxidation and carbohydrate metabolism, e.g. alanine, while the quantities of glutamine and asparagine decrease (Monselise 1966, in Szotoryi et al. 1971).

The respiration intensity of irradiated fruit varieties with the stage of ripening: after the radiation treatment the intensity of respiration increases considerably in green or unripe fruits, while in ripe ones it is hardly perceptible.

Table 4

*Changes in the respiration of pears in the first three days after irradiation (averaged over 3 years)*

Variety	Treatment	1st day	2nd day	3rd day	Average
Hardenpont	Ø	510	528	490	510
téli vajkörte	1000 Gy	733	738	705	725
Serres	Ø	412	408	520	447
Olivér	100 Gy	573	586	631	597
LSD <sub>5%</sub>					359

Note: respiration CO<sub>2</sub> mg/24 h/kg fruit

*of storage in response to gamma radiation*

1983			1984			
2nd day	3rd day	Average	1st day	2nd day	3rd day	Average
651	637	641	460	506	419	462
781	777	765	804	784	707	765
427	448	439	371	405	659	478
713	680	702	423	454	803	560
433	387	418				
613	563	601				
401	375	401				
678	624	652				
37		21		46		27

This could be demonstrated primarily for pears and other fruit with climacteric respiration, and was much less pronounced for oranges and lemons.

Smock et al. (1957) and Romani et al. (1961) found that the intensity of respiration increased after irradiation but only for a few days. The intensity of respiration was much lower at the climacteric respiration peak than before the peak of the respiration curve. According to Maxie, Sommer, Muller and Rae (1966) at the beginning of climacteric respiration the intensity of respiration of irradiated Williams pears rose and the ethylene production increased, but the ripening of the fruit became irregular. On the other hand, after the climacteric respiration phase the slight increase in the intensity of respiration and in the ethylene production did not influence the ripening process.

In experiments carried out by Maxie, Eaks, Sommer, Rae and El-Batal (1964) it was found that at the beginning of the climacteric respiration period the respiration intensity of irradiated peach varieties of the nectarine type increased, as did the ethylene production, which, in turn, had a positive effect on fruit ripening.

On the other hand, Maxie (1963) observed that in fruit with non-climacteric respiration (e.g. oranges, grapefruits) the intensity of respiration rose immediately after the radiation treatment. Later Maxie et al. (1965) reported that gamma radiation increased the ethylene production in fruit with non-climacteric (e.g. lemon) and climacteric respiration alike.

Even today very little is really known as to the reasons for the increase in the intensity of respiration and the production of ethylene gas under the influence of radiation. Young (1965) is of the opinion that while radiation has no direct effect on the ethylene production of the fruit, its indirect effect on the physiological processes of the fruit is unfavourable. According to this author among the products of anaerobic respiration, alcohol and acetaldehyde



**Table 5**  
*Change in the respiration of winter pears*

Variety	Treatment	1982			
		A	B	C	
				*	**
Hardenpont	Ø	426	738	773	789
téli vajkörte	1000 Gy	646	792	805	778
Serres	Ø	341	504	729	684
Olivér	1000 Gy	610	662	959	789
Téli	Ø	353	518	611	583
esperes	1000 Gy	563	659	789	601
Nemes	Ø	648	587	915	673
Krasszán	1000 Gy	554	727	603	728
LSD <sub>5%</sub>		31	36	55	27

Note: respiration CO<sub>2</sub> mg/24 h/kg fruit

\* = Storage in unchanged atmosphere.

\*\* = Storage in regulated atmosphere.

accumulate to a greater extent in the tissues of irradiated fruit; in other cases the skin or peel may suffer radiation injury, as happens with citrus fruit. Then, as a consequence of the injury, the etheric oils oxidize more intensely and come into contact with enzymes.

### Materials and methods

The material of the experiment consisted of "Jonathan" apples and four pear varieties: "Nemes Krasszán", "Hardenpont téli vajkörte", "Téli esperes" and "Serres Olivér".

The experiments were coordinated at the Fruit production Department of the University of Horticulture and Food Industry. The gamma radiation treatments were carried out in the Isotope Institute of the Hungarian Academy of Sciences, and the X-ray treatments at the Multiradiation Department of the "Március 15" Cooperative Farm, Hernád.

Gamma radiation was applied at 500–1000–1500 Gy, and X-rays at 40–60–100 Gy.

The control and the irradiated fruit were stored for shorter or longer periods in the experimental cold-store of the University of Horticulture and Food Industry at Szigetsép, under constant or controlled atmosphere conditions.

The respiration of the apples and pears was measured on 3 occasions.

The first measurement was carried out after the radiation treatment (when putting the fruit into storage), the second in the middle of the storage period, and the third at the end of storage. Respiration was measured according to Hámori (in: Hámori-Sass 1971) and Bubán (1971). For each variety 2–3 fruit — depending on the size — were placed in a closed space at 25 °C. The amount of CO<sub>2</sub> produced in 16 hours was absorbed in 0.1 N NaOH. The cubic capacity of the measuring vessel was so chosen as to contain air and fruit at a ratio of 1 : 7 to 1 : 4.

According to Wilkinson (1961) this volumetric ratio did not influence the respiration of the fruit (as was confirmed by experiments at the department). The intensity of respiration was expressed as the quantity of carbon dioxide (mg) per unit weight of fruit (kg), or per unit time (24 hours).

Respiration was measured on three successive days, with 3 replications, at both the beginning and end of storage. On each occasion a measuring vessel without fruit was also used.

The experimental data were processed at the Plant Productions and Qualifications Institute, under the guidance of Péter Wellisch, mathematician.

*in response to gamma radiation*

1983				1984			
A	B	C		A	B	C	
		*	**			*	**
641	747	717	733	462	762	813	813
864	810	708	808	765	779	843	801
439	739	687	813	560	618	1029	806
702	779	800	760	478	839	992	877
418	547	652	702				
676	671	726					
401	605	614	758				
652	775	815	675				
21	44	27	36	27	18	50	34

**Results***Apples*

Respiration is a process characteristic of the ripening stage of the apple, so the changes brought about by various rates and types of radiation are of considerable importance. In the present experiment both X-rays and gamma radiation substantially increased the intensity of respiration at the beginning of storage, the increase was significant at the  $P = 5\%$  level (Tables 1-2).

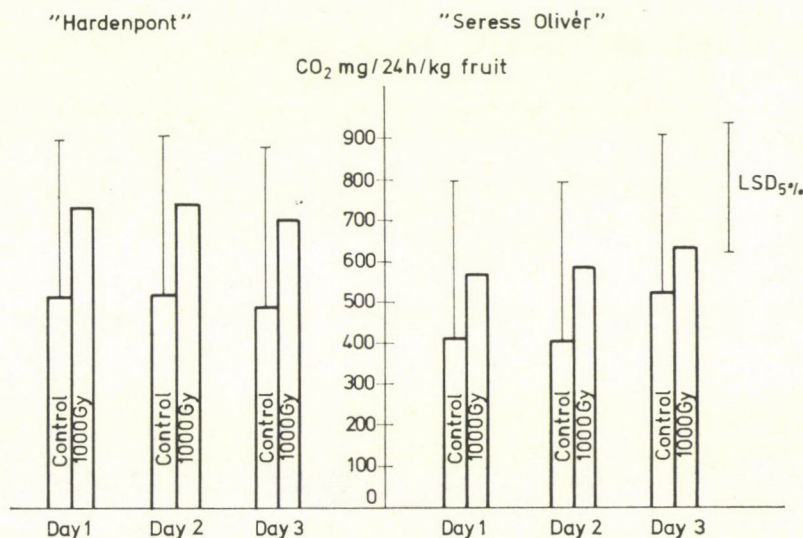


Fig. 3. Changes in the respiration of pears in the first few days following gamma radiation (3-year average)



"Hardenpont téli vajkörte"

"Seress Olivér"

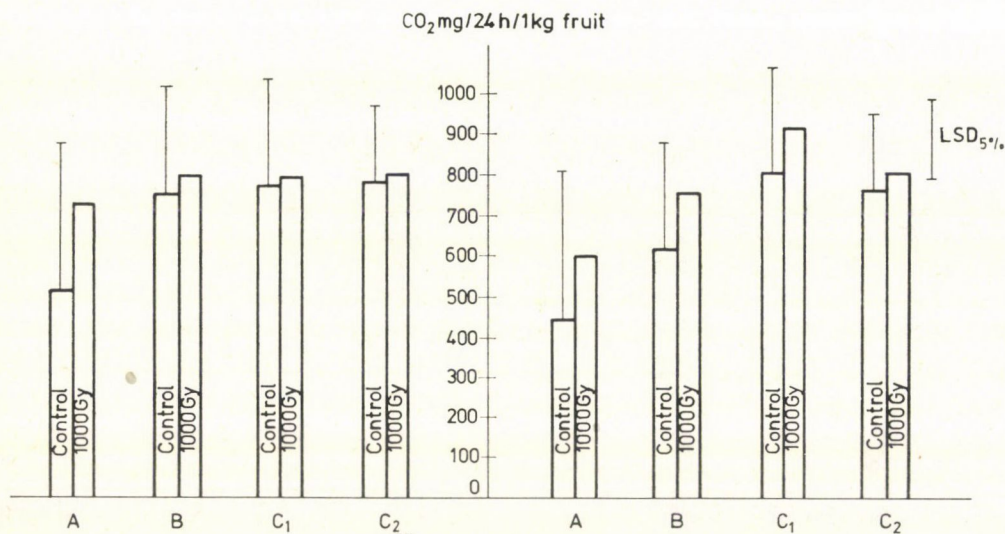


Fig. 4. Changes in the respiration of pears during storage in response to gamma radiation (3-years average).

A = at beginning of storage

B = in mid-storage

C<sub>1</sub> = at end of storage constant atmosphere

C<sub>2</sub> = at and of storage controlled atmosphere

For the sake of clarity, these results have also been illustrated graphically (Figs 1–2). The rapid initial increase in the intensity of respiration changed to a regular decrease after several days of storage, as was earlier proved by Smock et al. (1957) and Romani et al. (1961). This tendency could still be observed at the end of the storage period, so much so that apart from a few cases the respiration rate of irradiated fruit was then somewhat lower than that of the control.

Table 6

Changes in the respiration of pears in the course of storage in response to gamma radiation (averaged over 3 years)

Variety	Treatment	A	B	C	
				*	**
Hardenpont téli vajkörte	Ø	510	749	767	779
	1000 Gy	725	794	785	795
Serres Olivér	Ø	447	621	815	768
	1000 Gy	597	760	917	809
LSD <sub>5%</sub>		359	261	263	186

Note: respiration CO<sub>2</sub> mg/24 h/kg fruit

\* = Storage in unchanged atmosphere.

\*\* = Storage in regulated atmosphere.



## Pears

Great importance is attached to changes in the respiration values, since they express most markedly the physiological changes, the processes taking place during ripening.

Figures 3 and 4 were compiled using the results of a 3-year experimental series. They prove quite clearly the increase in respiration under the influence of gamma radiation, though the differences between the treatments are not significant. The increase is particularly remarkable at the beginning of the storage period (Tables 3 and 4), but there are measurable differences in the middle and at the end of the storage period as well (Table 5).

In Table 6 the effect of both gamma radiation and X-ray treatments can be seen. The role of the latter is less important. The differences obtained (10–30% depending on the variety) were mostly statistically significant.

## References

- Bubán T. (1971): Adatok az alma gyümölcs érettségi fokának meghatározásához (Determination of the ripening stage of apples). *Kertgazdaság*, **3**, 2, 68–71.
- Hátori-Szabó, J., Sass, P. (1971): A Jonathan alma érettségét jelző néhány tényező vizsgálata (Some factors indicating the ripeness of Jonathan apples). *Kertészeti Egyetem Közleményei*, **35**, 3, 191–201.
- Al Bachir, M., Sass, P. (1983): Gamma-sugarak hatásának vizsgálata kertészeti termények tárolása során. Sugárzási módszerek alkalmazása a mezőgazdaságban és az élelmiszeriparban (Effect of gamma radiation in the course of storing horticultural produce. Application of radiological methods in agriculture and the food industry). *Atomki riport*, **10**, 5, 176–178.
- Al Bachir, M., Sass, P. (1983): Study on the effect of gamma radiation in the course of storing horticultural produce. (In: Utilization of radiation energy in the food industry and in agriculture). *Central Food Research Institute, Budapest*, 15–19.
- Al Bachir, M., Sass, P. (1984): Besugárzott gyümölcsök tárolás alatti változásai (Changes in irradiated fruit during storage). *Kertészeti Egyetem Közleményei*, Budapest (in press).
- Al Bachir, M., Várady-Burgett C. (1986): A röntgensugárzás hatása a Magyar kajszli színeződésére (Effect of X-ray treatments on fruit colouring in the apricot variety Magyar kajszli). *Mérés és Automatika*, Budapest (in press).
- Maxie, E. C., Sommer, N. F., Brown, D. S. (1964): Radiation technology in conjunction with post-harvest procedures as a means of extending the shelf-life of fruits and vegetables. *US Atomic Energy Commission, Research and Develop.* **34**, 80–86.
- Maxie, E. C., Eaks, I. L., Sommer, N. F., Rae, H. L., El-Batal, S. (1965): Effect of gamma radiation on rate of ethylene and carbon dioxide evolution by lemon fruit. *Plant Physiol.* **40**, 407–410.
- Maxie, E. C., Sommer, N. F., Muller, C. J., Rae, H. L. (1966): Effect of gamma radiation on the ripening of Bartlett pears. *Plant Physiol.* **41**, 437–440.
- Metlitsky, L. V. (1975): *A gyümölcsök és zöldségek biokémiája*. (Biochemistry of fruit and vegetables) Mezőgazdasági Kiadó, Budapest.
- Sass, P. (1986): *Gyümölcstárolás* (Fruit storage). Mezőgazdasági Kiadó, Budapest.
- Szotyori, K. L., Lindner, K., Andrassy, É., Alexis, H. (1971): Kis dózisú gamma-sugárzás hatás a citrom utóérésére (Effect of low rate gamma radiation on postripening in lemons). *Élelmiszer Vizsgálati Közlemények* **17**.
- Wilkinson, B. G. (1981): A simple method for following climacteric respiration in apples. *J. Hort. Sci.* **36**, 197–201.
- Young, R. E. (1965): Effect of ionizing radiation on respiration and ethylene production of avocado fruit. *Nature*, London. **205**, 1113.





## TOLERANCE OF FIVE OIL CROPS TO SALINITY AND TEMPERATURE STRESSES DURING GERMINATION

F. S. EL NAKHLAWY and M. A. EL FAWAL

AGRONOMY AND RANGE SCIENCE DEPARTMENT, COLLEGE OF AGRICULTURE,  
KING SAUD UNIVERSITY, QASSIM BRANCH, SAUDI ARABIA

(Received: 26 November 1986; accepted 26 August 1987)

This study aimed to elucidate the effects of sodium chloride and sodium sulphate using four concentrations (0.0, 5.000, 10.000 and 15.000 ppm) on germination percentage and seedling characteristics of five oil crops under three levels of day/night temperature (20/7, 25/12 and 30/17 °C). The five oil crops were sunflower (*Helianthus annuus* L.), safflower (*Carthamus tinctorius* L.), soybean (*Glycine max* (L.) Merr.), flax (*Linum usitatissimum* L.) and rape (*Brassica napus* L.). The results indicated that sodium chloride has more adverse effect on germination and seedling characteristics than does sodium sulphate. The percentage of germination and normal seedlings were reduced as the salinity concentration increased. For seed germination potentiality, flax ranked highest, followed by sunflower, safflower, soybean and rape. Sunflower and safflower were the most tolerant crops, while flax and soybean were the least, in terms of normal seedling growth under the different temperature and salinity treatments.

**Keywords:** *Helianthus annuus* L., *Carthamus tinctorius* L., *Glycine max* L., *Linum usitatissimum* L., *Brassica napus* L., salinity and temperature stress, seed germination

### Introduction

In arid and semi-arid countries the agricultural development confronts such environmental challenges as salinity, temperature and drought stresses. Water potential and temperature are important environmental factors affecting seed germination. The ability of seeds to germinate and emerge under salt stress indicates that they may have the genetic potential for salt tolerance, at least at an early stage of the life cycle (Pearson and Bernstein 1959 and Pearson et al. 1966).

Dewey (1962) has suggested that germination provides a fair measure of general salt tolerance. However, Ayers and Hayward (1948) stated that positive relationship was found between salinity tolerance during germination and tolerance at other development stages. Donovan and Day (1969) noted that germination is affected by both salt concentration (the osmotic pressure) and the type of salt. Kaufmann and Ross (1970) concluded that the effect of water stress at one temperature is not valid at another temperature, if a water stress-temperature interaction exists.

In Saudi Arabia, where the production of oil crops is very low, some main oil crops may be exploited, provided that well adapted cultivars are selected. The main objective of the present study is to evaluate the tolerance to salinity and temperature stresses of five oil crops during their germination.



## Materials and methods

The seed germination of 5 oil crops was studied under different treatments of saline concentration and temperature. The oil crops were sunflower (*Helianthus annuus* L.), safflower (*Carthamus tinctorius* L.), soybean (*Glycine max* (L.) Merr.), flax (*Linum usitatissimum* L.), and rape (*Brassica napus* L.). Two salts, namely sodium chloride (NaCl) and sodium sulphate ( $\text{Na}_2\text{SO}_4$ ), each with 4 concentrations (0, 5.000, 10.000 and 15.000 ppm), were used. The 3 temperature treatments were alternating 20/7 °C, 25/12 °C and 30/17 °C, and the duration of the alternating temperature was 12 hours. Seeds were germinated on filter paper in sterilized Petri dishes, each dish contained 30 seeds and each treatment was replicated 3 times. The experimental design was split-split-split plot. The main treatments, sub-treatments, sub-sub-treatments and sub-sub-sub treatments were temperatures, salts, concentrations and oil crops, respectively. The traits studied were germination percentage, number of normal seedlings and seedling length. The arcsine square root transformation of germination percentage was used in the analysis of variance, to stabilize the treatment variances (Steel and torrie 1984). Germination counts growth measurements were determined after 7 days of inhibition.

## Results and discussion

### *Germination percentage*

Generally, the present results proved that the studied oil crops greatly differed with respect to the tolerance of the environmental factors affecting seed germination (Tables 1, 2, 3 and 4). These crops may be ranked according to the potentiality of seed germination; flax was the highest followed by sunflower, safflower and soybean, while rape was the lowest. For the influence of temperature treatments, the medium level (25/12 °C) gave the highest germination percentage for all crops except flax, for which the highest germination percentage was obtained at the lowest level (20/7 °C). Both flax and sunflower ranked highest under all temperature treatments, rape was the lowest.

With respect to salt treatments, seed germination rates were generally reduced by different solutions. The influence of sodium chloride on germination was clearly higher than that of sodium sulphate. Similar results were obtained by Flowers et al. (1977). All crops responded variously to the salt treatments. Sunflower and flax were the most tolerant, followed by safflower soybean and rape under the different temperature levels. In regard to the treatments of sodium chloride concentrations, the rates of seed germination for all crops tended to decrease with the higher concentration, especially under the lowest and highest temperature levels (20/7 °C and 30/17 °C). These results are confirmed by those obtained by Islam (1978) on soybean, Gaur and Tomar (1975) on sunflower, Rizk et al. (1979) on rape and Goswami et al. (1978) on safflower. It is worth mentioning that greater NaCl concentrations resulted in higher germination rates in the case of sunflower, under the medium and high levels of temperature. For flax, the same germination percentages obtained up to 10.000 ppm. For safflower and soybean 5.000 ppm solution gave a slight stimulation for such treatments. As for the concentration levels of

Table 1

*Results of significance tests for the effect of temperature, salt and salt concentration on germination and seedling characteristics of five oil crops*

Source of variation	D.F.	Germination, %	No. of normal seedlings	Seedling length
<i>Main plots</i>				
Blocks	2			
Temperatures (T)	2	**	**	*
Error "a"	4			
<i>Sub-plots</i>				
Salts	1	**	**	**
TXS	2	NS	NS	NS
Errors "b"	6			
<i>Sub-sub-plots</i>				
Concentrations (C)	3	**	**	**
TXC	6	NS	*	**
SXC	3	**	**	**
TXSXC	6	NS	*	NS
Error "C"	36			
<i>Sub-sub-plots</i>				
Oil crops (0)	4	**	**	**
TXO	8	*	**	**
SXO	4	*	**	**
CXO	12	*	**	**
TXSXO	8	NS	**	**
TXCXO	24	NS	NS	**
TXCXO	12	*	**	**
TXSXCXO	24	NS	*	**
Error "d"	192			
Total	359			

\* Significant at the 0.05% level of probability.

\*\* Significant at the 0.01% level of probability.

NS Non-significant.

sodium sulphate, the higher concentrations were accompanied by reductions in germination rate under the lower level of temperature, except in the case of flax. At the highest salt concentration (15,000 ppm), the response of all crops under the 3 levels of temperature was obviously different in terms of germination percentage.

### *Seedling characteristics*

The number of normal seedlings and their length were determined. Since these oil crops are genetically different with respect to the seedling characteristics, the data in Table 3 represent the ratio between the different treatments and the control in percentages.



*Number of normal seedlings*

The present results indicated the differences among the 5 oil crops, regarding the growth of seedlings under different temperatures and salinity treatments. Generally speaking, sunflower produced the greatest percentage of normal seedlings, followed by safflower, rape, flax and soybean. The present study showed that the median level of temperature (25/12 °C) stimulated the normal growth of seedlings for all crops except soybean, in which the greatest percentage of normal seedlings was obtained under the highest temperature (30/17 °C). The present results are similar to those obtained by Tyagi and Tripathi (1983), Matthews, Hayes (1982) and Jordan (1975) on soybean. They found that the optimum temperature for both the germination and emergence of soybean was 20 °C to 28 °C. Cserenyés (1973) showed that sunflower is sensitive to salinity at 10 °C and the optimum was obtained between 25 and 30 °C.

Comparing the salt influences, the percentages, of normal seedlings treated with NaCl were lower than those treated with Na<sub>2</sub>SO<sub>4</sub> for all crops. The percentage of normal seedlings was obviously reduced as the concentration

Table 2

*Germination percentage of the crops as influenced by concentrations of NaCl and Na<sub>2</sub>SO<sub>4</sub> under the levels of day/night temperature*

Temperature, °C	Salt	Salt concentration (ppm)	Germination (%)				
			Sunflower	Safflower	Soybean	Flax	Rape
20/7	NaCl	0.0	93.3	93.3	76.7	100	80
		5,000	90	83.3	76.7	90	70
		10,000	80	83.3	46.7	80	46.7
		15,000	80	70	26.7	13.3	3.3
	Na <sub>2</sub> SO <sub>4</sub>	0.0	90	80	83.3	100	90
		5,000	83.3	80	66.7	100	66.7
		10,000	80	83.3	76.7	96.7	63.3
		15,000	76.7	76.7	76.7	100	46.7
	NaCl	0.0	80	90	100	96.7	90
		5,000	86.7	86.7	76.7	96.7	76.7
		10,000	86.7	76.7	70	96.7	56.7
		15,000	90	76.7	56.7	33.7	20
	Na <sub>2</sub> SO <sub>4</sub>	0.0	83.3	80	86.7	96.7	86.7
		5,000	90	86.7	86.7	86.7	80
		10,000	90	86.7	96.7	93.3	73.3
		15,000	90	80	73.3	93.3	80
25/12	NaCl	0.0	80	90	100	96.7	90
		5,000	86.7	86.7	76.7	96.7	76.7
		10,000	86.7	76.7	70	96.7	56.7
		15,000	90	76.7	56.7	33.7	20
	Na <sub>2</sub> SO <sub>4</sub>	0.0	83.3	80	86.7	96.7	86.7
		5,000	90	86.7	86.7	86.7	80
		10,000	90	86.7	96.7	93.3	73.3
		15,000	90	80	73.3	93.3	80
	NaCl	0.0	76.7	80	80	100	66.7
		5,000	86.7	83.3	83.3	96.7	46.7
		10,000	90	63.3	40	96.7	43.3
		15,000	70	70	30	16.7	30
	Na <sub>2</sub> SO <sub>4</sub>	0.0	86.7	80	66.7	96.7	43.3
		5,000	70	86.7	66.7	96.7	66.7
		10,000	86.7	66.7	80	96.7	36.7
		15,000	73.3	70	66.7	96.7	40
30/17	NaCl	0.0	76.7	80	80	100	66.7
		5,000	86.7	83.3	83.3	96.7	46.7
		10,000	90	63.3	40	96.7	43.3
		15,000	70	70	30	16.7	30
	Na <sub>2</sub> SO <sub>4</sub>	0.0	86.7	80	66.7	96.7	43.3
		5,000	70	86.7	66.7	96.7	66.7
		10,000	86.7	66.7	80	96.7	36.7
		15,000	73.3	70	66.7	96.7	40

Table 3

Effect of salt concentration and temperature on number of normal seedlings and seedling length expressed as percentage of their corresponding control (0.0 salt concentration)

Temperature °C	Salt	Salt concentration (ppm)	Normal seedlings					Seedling length				
			Sonflower	Safflower	Soybean	Flax	Rape	Sunflower	Safflower	Soybean	Flax	Rape
20/7	NaCl	0.0	100	100	100	100	100	100	100	100	100	100
		5,000	65.4	57.7	88.2	53.3	70.8	85.5	81.7	77.9	50.9	80.9
		10,000	3.8	53.8	5.8	30	41.7	43.3	80.4	0.00	14.2	49.6
		15,000	0.0	0.0	0.0	0.0	0.0	0.00	0.00	0.00	0.00	0.00
	Na <sub>2</sub> SO <sub>4</sub>	0.0	100	100	100	100	100	100	100	100	100	100
		5,000	90.5	84.9	83.3	100	71.4	125.5	122.9	100	63.9	100
		10,000	85.7	45	50	76.7	71.4	83.2	83.1	60.0	53.3	34.9
		15,000	0.0	26.7	42.8	0.0	42.8	0.0	70	24.1	0.0	12.2
	NaCl	0.0	100	100	100	100	100	100	100	100	100	100
		5,000	100	77.8	62.5	89.7	66.7	68.1	78.7	51.0	58.3	65.1
		10,000	100	66.7	37.5	82.7	61.9	72.3	54.1	32.6	32.6	40.3
		15,000	25	40.7	0.0	0.0	4.7	9	36.1	0.0	0.0	0.0
25/12	Na <sub>2</sub> SO <sub>4</sub>	0.0	100	100	100	100	100	100	100	100	100	100
		5,000	92	100	92.3	90	76.2	77.2	89.1	63.1	78.4	81.7
		10,000	88	75	84.6	60	66.7	36.0	87.3	51.2	31.6	32.2
		15,000	88	45	0.0	30	61.9	32.3	47.3	0.0	21.6	20.1
	NaCl	0.0	100	100	100	100	100	100	100	100	100	100
		5,000	91.6	87.5	77.8	86.7	61	90.3	78.6	55.3	67.2	93.2
		10,000	70.9	50	14.8	43.3	52.2	69.3	47.1	41.3	62.5	62.9
		15,000	0.0	0.0	0.0	0.0	26.1	0.0	0.0	0.0	0.0	16.7
	Na <sub>2</sub> SO <sub>4</sub>	0.0	100	100	100	100	100	100	100	100	100	100
		5,000	65.3	75	80.8	96.7	73.9	94	91	73.3	95.3	100
		10,000	57.7	45.9	76.9	50	43.4	66.5	48.9	64.3	34.7	17.7
		15,000	34.6	45	50	0.0	34.8	22.9	26.8	45.5	0.0	12.1



Table 4

*LSD test at 0.05 for germination (%), normal seedlings and seedling length*

Treatments	Germination, %	No. of normal seedlings	Seedling length
Temperatures (T)	1.92	4.10	3.99
SALT (S)	3.94	1.93	4.50
Concentrations (C)	3.30	3.10	4.00
Oil crops (O)	3.67	3.40	4.70
TXC	5.72	5.30	6.87
SXC	4.67	4.40	5.42
TXO	3.67	3.40	4.70
SXO	6.36	5.95	6.12
CXO	7.35	4.86	6.63
TXSXO	9.00	8.41	10.38
TXCXO	12.70	11.90	16.20
SXCXO	10.40	9.71	13.30
TXSXCXO	18.00	16.80	23.00

of salinity increased. At the higher concentration (15,000 ppm) of NaCl all the crops produced abnormal seedlings under both low and high temperature levels (20/7 and 30/17 °C), except rape under 30/17 °C. Under the median temperature level (25/12 °C), however, the percentage of normal seedlings of sunflower and flax was clearly high at a concentration of 10,000 ppm. Except in a few individual cases, all crops showed a reasonable tolerance to the high concentrations of Na<sub>2</sub>SO<sub>4</sub>, especially under the median level of temperature (25/12 °C).

### *Seedling length*

These oil crops varied with respect to the influence of studied treatments on seedling length. Safflower ranked highest in the tolerance to temperature and salinity in terms of normal seedling growth while flax and soybean had the highest sensitivity. Under the temperature treatments, the seedling length was relatively better at the lower and higher levels, 20/7 °C and 30/17 °C respectively.

The present study indicates that the growth of seedlings was severely affected by NaCl rather than by Na<sub>2</sub>SO<sub>4</sub>, in all cases. With different concentrations of salinity, the growth of seedlings obviously declined as the NaCl concentration increased. On the other hand, A 5,000 ppm concentration of Na<sub>2</sub>SO<sub>4</sub> stimulated the seedling growth in the case of both sunflower and safflower under the lowest level of temperature.

## Acknowledgement

The authors would like to acknowledge the assistance of Mr. M. Basyoni, laboratory technician.

## References

- Ayers, A. D., Hayward, H. E. (1948): A method for measuring the effects of soil salinity on seed germination with observations on several crop plants. *Soil Sci. Soc. Amer. Proc.* **13** 224-226.
- Cseresnyes, Z. (1979): The germination of *Helianthus annuus* seeds under optimum laboratory conditions. *Seed Sci. and Technol.* **7**, 319-328.
- Dewey, D. R. (1962): Germination of crested wheat grass in salinized soil. *Agron. J.* **54**, 353-355.
- Donovan, J. T., Day, A. D. (1969): Some effects of high salinity on germination and emergence of barley. *Agron. J.* **61**, 236-238.
- Flowers, T. J., Troke, P. F., Yeo, A. R. (1977): The mechanism of salt tolerance in halophytes. *Ann. Rev. Plant Physiol.*, **28**, 89-121.
- Gaur, B. L., Tomar, D. S. (1975): Effect of salinity on the germination of sunflower (*Helianthus annuus* L.) varieties. *Science and Culture*, **41**, 429-430.
- Goswami, S. K., Gehlot, C. L., Lal, K. (1978): A note on salt tolerance of different varieties of safflower (*Carthamus tinctorius* L.) at germination. *Madras Agric. J.* **65**, 137-138.
- Islam, M. T. (1978): Effects of sodium chloride on soybean seed germination. *Bangladesh J. Bot.* **7**, 107-109.
- Jordan, C. W. (1975): Some effects of temperature on germination of soybeans (*Glycine max*) as related to cultivar and seed vigor. *Diss. Abst. Int.*, B. **63**, 3, 1015.
- Kaufmann, M. R., Ress, K. J. (1970): Water potential, temperature and kinetin effects on seed germination in soil and solute systems. *Am. J. Bot.* **57**, 413-419.
- Matthews, D. J., Hayes, P. (1982): Effect of temperature on germination and emergence of six cultivars of soybean (*Glycine max*). *Seed Sci. et Technol.*, **10**, 547-555.
- Pearson, G. A., Ayers, A. D., Eberhard, D. L. (1966): Relative salt tolerance of rice during germination and early seedling development. *Soil Sci.*, **102**, 151-156.
- Pearson, G. A., Bernstein, L. (1959): Salinity effects at several growth stages of rice. *Agron. J.*, **51**, 654-657.
- Rizk, T. Y., Ali, H. A., Al-Hasan, A. M. (1979): Effect of varying concentrations of certain salts on germination and seedling vigour of two rape (*Brassica napus* L.) varieties. *Mesopotamia J. Agric.*, **14**, 25-40.
- Steel, R. G. O., Torrie, J. H. (1980): *Principle and procedures of statistics*. McGraw-Hill Book Company Inc., N. Y.
- Tyagi, S. K., Tripathi, R. P. (1983): Effect of temperature on soybean germination. *Plant and Soil* **74**, 273-280.





## EFFECT OF KINETIN AND SALINITY ON OSMOTIC PRESSURE AND CARBOHYDRATE CONTENTS IN TWO CROP PLANTS

F. M. SALAMA and A. A. AWADALLA

BOTANY DEPARTMENT, FACULTY OF SCIENCE, ASSIUT UNIVERSITY, ASSIUT, EGYPT

(Received: 25 January 1987; accepted 16 April 1987)

Increase in foliar osmotic potential to soil osmolality has been previously realized by the senior author and some other investigators. The present study has been carried out to clarify the interactive kinetin and salinity stress on osmotic potential and carbohydrate contents in cotton and millet plants. Kinetin solution (10 ppm) was applied in three different modes; presoaking of seeds, spraying of shoot system and with irrigation water. This study revealed that kinetin increased the osmotic pressure in the two experimental plants when applied by spraying and irrigation with water. However the osmotic pressure was reduced when the seeds were presoaked in kinetin solution. It was concluded that kinetin can alleviate the effect of salinity stress on the osmotic pressure. The carbohydrate contents of various treated plants were also taken into consideration.

**Keywords:** *Gossypium barbadense* Cv. "Dandara", *Sorghum bicolor* L. Cv. "Giza-3", kinetin, osmotic pressure, carbohydrate contents

### Introduction

Crop plants may appear (ecological) to endure increased soil salinity but there usually occurs some internal (physiological) disorders. These disorders are particularly in metabolic pathways which are generally reflected in decreased growth and productivity. The means by which plants resist reduced soil water potential, whether matric or osmotic, are classified into morphological (adaptation) and physiologic (adjustment). A detailed account of such means is provided by both Stocker (1960) and Parker (1969). Increased soil osmolality has been mostly accompanied by a parallel increase in foliar osmotic potential (Salama et al. 1980; El-Sharkawi and Salama 1973; 1976; Cooper and Dumbroof 1973; Simmelgaard 1976 and Oertli 1976). In this respect Pallas and Box (1970) observed that after treatment of excised leaves with kinetin, the osmotic potential becomes less negative. Similarly Tal and Imber (1971) found that kinetin decreased the osmotic pressure in tomato.

Similarly there is some evidence that soil moisture stress leads to a decrease in reserve (insoluble) carbohydrates and to an increase in soluble sugars (Stocker 1960; Vaadia et al. 1961; Parker 1969). Treatment with cytokinins produced some effects upon the physiology and metabolism of water-stressed plants. Alternatively, Mothes (1964) reported an accumulation of carbohydrates



within the detached leaves after treatment with kinetin. Some authors have reported that treatment with kinetin resulted in starch breakdown and in an increase in free sugars within the plant tissue (Dennis et al. 1967; Boothby and Wright 1962; Nevin et al. 1966; Street et al. 1970; Berridge and Ralph 1971). Obviously the interaction of kinetin and salinity stress sometimes provided contrasting results. This variation in results depends mainly on the plant type, and the level of stress, as well as on the type and concentration of the cytokinins used. Thus, the present work intends to investigate the interactive effect of kinetin (applied by three different methods), and salinity stress on osmotic pressure and carbohydrate contents in *Sorghum* and *Gossypium* plants.

### Materials and methods

The experimental plants, were cotton (*Gossypium barbadense* CV Dandara) and millets (*Sorghum bicolor* L. CV Giza-3). These plants were grown in plastic pots containing 1400 gm air dry soil (sand/clay 2 : 1 v/v), watered twice with 100 ml portions of full strength Hoagland solution (Hoagland and Arnon, 1950). Five plants were allowed to grow in each pot,  $\psi_s$  levels were chosen at -3, -7, -10 and -13 bar, in addition to the control (-0.3 bar). For each potential level, 3 pots were assigned at random. An osmotic solution prepared according to the formula given by Lagerwerff and Eagle (1961) was used in irrigation to adjust  $\psi_s$  to the desired levels. A mixture of  $\text{CaCl}_2$  and  $\text{NaCl}$  was used in the preparation of this solution in which the sodium adsorption ratio was fixed at 12.5%. Solutions were added to the soil in such a way that the soil solution acquired the assigned potential at field capacity. Treatments of plants with saline solutions began when seedlings were 8 weeks old (except in the presoaking experiment). On completing the treatment, the plants were watered with distilled water only. The moisture content of the soil was never allowed to fall below field capacity. This was achieved by checking weights of pots twice daily. The plants were allowed to be subjected to treatment for a period of 2 weeks before treatment with kinetin solution. The kinetin concentration in the water solution used throughout the experiment was 10 ppm. Three different methods of kinetin application have been used in the present study, namely presoaking of seeds, spraying of shoot system and with irrigation water. In the presoaking method seeds of experimental plants were soaked in kinetin solution for 8 hours, then air dried for 24 hours. The dried seeds were soaked again in the kinetin solution for another 8 hours and then dried for 24 hours. The treated seeds were then dried for 24 hours. The treated seeds were then sown in salinized pots containing 1400 gm soil with the different levels of  $\psi_s$  previously mentioned. Another group of seeds were soaked in the same manner but in distilled water and were treated similarly for comparison between then and the seeds treated with kinetin. In the spraying method, kinetin solution was applied by spraying the shoot system of the growing plants in each pot with 10 ml of hormone solution. Control plants were sprayed with distilled water. Reapplication with kinetin was performed 5 days after the first spraying. The measurements were recorded 7 days after spraying with the second dose. In the irrigation method, each pot was irrigated with a total of 200 ml of kinetin solution at intervals during a week. The measurements were recorded 7 days after the last irrigation, in which the soil water content was completed to field capacity by distilled water throughout the entire experiment.

The osmotic pressure of leaf sap was measured by the cryoscopic method (Walter, 1949) and details on this were described by El-Sharkawi and Abdel Rahman (1974). The partial osmotic pressure due to the ionic fraction was calculated from the equation (Black et al. 1965):

$$\text{Op (atm.)} = \text{Ec (in mmhos)} \times 0.36$$

The osmotic pressure values measured by the cryoscopic method are referred to as the "total" osmotic material whereas the partial osmotic pressure calculated from conductivity data is the "ionic" fraction. Free sugars were estimated in the leaf sap by the phenolsulphuric acid procedure (Dubois et al. 1956). Total hydrolysable (reserve) saccharides were determined by hydrolysable (reserve) saccharides were determined by hydrolysing aliquot samples of the



plant extract in 0.7 N hydrochloric acid (Pucher et al., 1957) and free sugars estimated in the hydrolysates as mentioned above. Proper statistical tests were used to elucidate the effects of single factor ( $\psi_s$  and kinetin) as well as their interaction. The tests included analysis of variance (F values) and least significant difference test LSD (to test for critical levels of effect of single factor). In case of significant effect of any of the single factors or interaction, the relative contribution (share) of such factors in the total response to treatment combination are

$$\eta^2 = \frac{\text{sum of squares due to the factor}}{\text{total sum of squares due to the treatment combinations}}$$

This coefficient is expressed as a fraction or a percentage. Such biometrical tests are applied according to the procedures of Ostle (1963) and Ploxinski (1969).

## Results

### *Application of kinetin by spraying*

The response of leaf osmotic concentration in the 2 experimental plants to reduced  $\psi_s$  is shown in Tables 1 and 2 for *Sorghum* and *Gossypium* respectively. The total osmotic pressure in the unsprayed plants with kinetin increased in both plants with increasing soil salinity. Spraying with kinetin solution nearly counteracted the effect of salinity and resulted in non-significant increases in osmotic pressure as compared with that of plants subjected to the corresponding salinized levels. The ionic fraction of osmotic pressure in unsprayed plants was relatively low, reaching only 1/10 of the total osmotic material in the control plants in *Sorghum* and 1/20 in *Gossypium*. The ionic fraction in sprayed plants increased significantly in both experimental plants. Analysis of variance of the data of osmotic pressure of either *Sorghum* or *Gossypium* indicated that salinity stress significantly affected the osmotic pressure in both plants (Table 3).

The results of sap analysis for both soluble and reserve carbohydrates are shown in Table 1 for *Sorghum* and Table 2 for *Gossypium*. In unsprayed *Sorghum* plants the soluble carbohydrates generally decreased with reducing  $\psi_s$  and the reduction was highly significant at -7 and -10 bar. On the other hand, the reserve sugars increased with reducing  $\psi_s$  and this increase was highly significant at -3 and -10 bar as compared with control plants. The increase in the soluble sugars due to spraying with kinetin solution was only highly significant at non-stressed plants and at -10 bar. The decrease in the reserve carbohydrates was significant in non-stressed plants, -3, -7 and highly significant in plants subjected to -10 bar as compared with those subjected to the corresponding salinity levels. In *Gossypium* (Table 2), the contents of the soluble sugars in plants, either sprayed or not with kinetin solution, decreased significantly with reducing  $\psi_s$ , while the reverse occurred in the case of reserve carbohydrates. The decrease in soluble sugars may be attributed to the water deficiency in the leaves which cause a reduction in the photosyn-



Table 1

Mean values of osmotic pressure (in atmospheres), soluble sugars (SS) and reserve sugars (RS) (mg./g./lf.d.wt.) in leaf sap of Sorghum at different levels of soil salinity, treated or not with kinetin solution applied by three different methods

App.		Spraying				Irrigating				Presoaking			
Kin	$\psi_s$ (bar)	OP		Sugars		OP		Sugars		OP		Sugars	
		Total	Ionic	SS	RS	Total	Ionic	SS	RS	Total	Ionic	SS	RS
Not treated	0	10.6	0.50	68.3	30.0	10.3	0.14	96.7	53.0	17.3	0.77	106.7	56.7
	-3	12.8	0.50	65.0	36.7**	21.2	0.54	88.3	68.3**	32.5**	1.60	86.7**	76.7**
	-7	15.9	0.63	51.2**	33.3	17.1	0.70	21.7**	61.7**	33.8**	1.90	48.3**	95.0**
	-10	18.3*	0.65	31.7**	43.3**	21.6	2.60*	30.0**	48.3	—	—	—	—
	-13	—	—	—	—	23.6*	1.90	16.7**	58.3	—	—	—	—
LSD	1‰	7.8	0.48	15.0	6.10	18.2	3.40	14.2	14.6	6.0	1.20	8.7	10.0
	5‰	5.8	0.33	10.3	4.20	12.5	2.30	9.1	10.1	4.1	0.48	5.9	6.9
Treated	0	13.7	0.60	85.0**	23.3*	8.8	0.14	108.3	51.7	11.2**	0.34	108.3	50.0
	-3	14.4	0.51	74.0	28.3*	19.9	0.64	101.7*	51.7**	18.3**	0.77	101.7*	55.0*
	-7	15.9	0.88	58.3	26.7*	15.1	0.30	40.0**	63.3	27.4**	2.10	78.3**	65.0**
	-10	18.2	0.83	53.3**	23.3**	23.2	1.50	17.7*	45.0	31.1	2.10	48.3	58.3
	-13	—	—	—	—	34.2*	3.30	15.0	63.3	—	—	—	—
LSD	1‰	4.9	0.60	16.3	9.4	15.1	3.50	17.2	14.2	2.8	1.20	20.8	24.0
	5‰	3.4	0.41	11.2	6.4	10.4	2.40	11.8	9.7	1.9	0.84	14.3	16.5

\* Significant  $P < 0.05$ .

\*\* Significant  $P < 0.01$ .

Table 2

Mean values of osmotic pressure (in atmospheres), soluble sugars (SS) and reserve sugars (RS) (mg.g.lf.d.wt.) in leaf sap of *Gossypium* at different levels of soil salinity, treated or not with kinetin solution applied by three different methods

App.		Spraying				Irrigating				Presoaking			
Kin.	$\psi_s$ (bar)	OP		Sugars		OP		Sugars		OP		Sugars	
		Total	Ionic	SS	RS	Total	Ionic	SS	RS	Total	Ionic	SS	RS
Not treated	0	17.4	0.37	53.30	21.7	16.8	0.50	40.0	23.3	18.6	0.53	41.7	20.0
	-3	20.7	0.98	41.70*	35.0*	20.9	0.96	31.7**	31.7	22.9	0.85	30.0**	23.3
	-7	23.2	2.10	25.00**	43.3**	23.9	1.20	21.7**	31.7	32.6**	2.00**	21.7**	26.7**
	-10	36.3**	1.50	20.00**	20.0**	21.2	1.60*	20.0**	21.7	—	—	—	—
	-13	33.7**	2.10	20.00**	35.0*	29.9**	1.80*	21.7**	21.0	—	—	—	—
LSD	1%	9.20	2.70	11.80	13.9	11.2	1.30	8.30	12.7	10.8	0.86	5.00	7.90
	5%	6.30	1.90	8.10	9.60	7.70	0.89	5.70	8.7	7.40	0.59	3.40	5.50
Treated	0	18.20	1.30	55.00	18.3	18.4	0.83	45.0	25.0	16.2	0.45	43.30	20.00
	-3	20.70	0.74	50.00*	26.7*	17.6	0.71	41.7*	21.7*	20.9	0.77	41.70*	16.70
	-7	23.00	1.90	35.00*	35.0*	17.6	0.63	40.0**	11.7**	24.1	1.10**	33.30**	10.00**
	-10	37.40	1.60	38.30**	26.7**	23.4	1.80	31.7**	10.0**	24.2	2.90	20.00	14.40
	-13	37.80	2.80	23.30	25.0	33.2	2.80	25.0	10.0**	—	—	—	—
LSD	1%	12.90	2.90	10.00	8.7	25.9	2.10	11.8	11.8	9.10	0.84	6.10	11.30
	5%	8.90	1.90	6.90	5.7	17.8	1.50	8.10	8.1	6.30	0.58	4.20	7.70

\* Significant  $P < 0.05$ .

\*\* Significant  $P < 0.01$ .



Table 3

*F* values and coefficient of determination,  $\eta^2$ , values for the significant effects of kinetin (applied by hydrates (soluble, SS and reserve, RS))

App.	Source of variance	Sorghum					
		Osmotic pressure				Carbohydrates	
		Total		Ionic		SS	
		F	$\eta^2$	F	$\eta^2$	F	$\eta^2$
Spraying	Kinetin	1.05	—	3.40	—	24.06**	0.04
	Salinity	50.50	0.98	20.90**	0.93	154.40**	0.94
	Kin $\times$ salinity	0.44	—	0.65	—	3.09*	0.02
Irrigating	Kinetin	0.31	—	0.06	—	7.45**	0.01
	Salinity	9.93**	0.89	6.09**	0.98	30.00**	0.97
	Kin $\times$ Salinity	1.16	—	1.25	—	6.84**	0.02
Presoaking	Kinetin	2.88	—	20.30**	0.02	98.50**	0.05
	Salinity	383.40**	0.64	186.8**	0.61	452.50**	0.90
	Kin $\times$ Salinity	213.00**	0.36	112.6**	0.37	22.60**	0.50

\*  $P < 0.05$ .

\*\*  $P < 0.01$ .

thetic activity. Spraying with kinetin solution increases soluble sugars significantly as compared with those of plants subjected only to the corresponding salinity levels. However the reverse was true with respect of reserve sugars. The analysis of variance of the data of carbohydrates of variously treated plants is shown in Table 3. Obviously in both plants, all *F* values for all factors (kinetin, salinity and their interaction) were significant with respect to either soluble or reserve sugars. The values of the determination coefficient  $\eta^2$  indicate that salinity stress had a dominator effect while both kinetin and its interaction had lesser values.

#### *Application of kinetin by irrigation*

The osmotic pressure of untreated water-stressed *Sorghum* plants increased with additional soil salinity but the increase was only significant at -13 bar, as compared with control plants (Table 1). Treatment with kinetin solution decreased the values of osmotic pressure until -7 bar; then it again increased, but this increase was significant only at -13 bar. The ionic fraction exhibits irregular behaviour; but the variations were of non-significant values except in *Sorghum* plants subjected to salinity of -10 bar, where the increase was significant.

In *Gossypium* (Table 2), the reduction in resulted in an increase in the total osmotic pressure of the leaf sap, but this increase was only significant

three different methods), salinity stress and their interaction, on osmotic pressure (OP) and carbohydrate of *Sorghum* and *Gossypium* plants

Gossypium									
RS		Osmotic pressure				Carbohydrates			
		Total		Ionic		SS		RS	
F	$\eta^2$	F	$\eta^2$	F	$\eta^2$	F	$\eta^2$	F	$\eta^2$
57.99**	0.10	0.83	—	1.80	—	38.77**	0.10	22.10**	0.19
129.80**	0.85	26.60**	0.98	2.40	—	78.80**	0.85	20.60**	0.72
8.80**	0.05	0.22	—	1.30	—	4.70**	0.05	2.60	—
2.51	—	0.19	—	0.49	—	59.60**	0.29	34.13**	0.38
10.00**	0.69	3.70*	0.87	6.47**	0.82	31.43**	0.62	8.94**	0.41
3.82*	0.27	0.49	—	1.31	—	4.50*	0.09	4.57*	0.21
0.01	—	20.30**	0.06	10.70**	0.04	93.11**	0.07	1.90	—
165.40**	0.77	59.20**	0.66	26.30**	0.47	602.20**	0.88	43.30**	0.70
49.80**	0.23	25.50**	0.28	32.40**	0.53	32.20**	0.05	17.70**	0.29

at -13 bar, as compared with that of control plants. Treatment with kinetin solution resulted in non-significant differences in the total osmotic pressure of the leaf sap of *Gossypium* compared with that of plants subjected to corresponding salinity levels. The ionic fraction represents about 5% of the total osmotic pressure in untreated plants. Treatment with kinetin solution tends to increase the ionic fraction in the leaf sap of these plants at all salinity levels except at -7 bar. Statistical analysis of the data of both experimental plants indicated that only salinity stress exerted significantly changing F values while kinetin and its interaction with salinity did not affect the osmotic pressure in the 2 experimental plants, since their F values were insignificant (Table 3).

Changes in carbohydrate contents of variously treated *Sorghum* and *Gossypium* plants are shown in Tables 1 and 2 respectively. The data indicate that generally the carbohydrate contents in *Sorghum* are higher than that in *Gossypium* plants. The soluble carbohydrate in stressed plants decreased significantly with reducing  $\psi_s$  in the 2 experimental plants as compared with those of plants subjected only to the corresponding salinity levels. Reserve sugars in untreated *Sorghum* plants increased with reducing and the increase was significant only at -3 bar, while in untreated *Gossypium* plants these sugars exhibited insignificant differences under the same conditions. Treatment of *Sorghum* plants with kinetin solution decreased the reserve carbohydrates significantly only at -3 bar, while in treated *Gossypium* plants, these



sugars decreased significantly at all salinity levels. The data in Table 3 show that all F values were significant in the two experimental plants except for the effect of kinetin on reserve sugars. The determination coefficient  $\eta^2$  indicates that the significant interaction shared with 2% and 9% in the case of soluble sugars and 27% and 21% in the case of reserve carbohydrates in *Sorghum* and *Gossypium* plants respectively.

#### *Application of kinetin by presoaking*

The osmotic concentration of leaf sap in untreated *Sorghum* plants increased significantly with reducing while the ionic fraction in the same plants increase non-significantly. Treatment with kinetin caused significant decreases in the total osmotic concentration at all levels in comparison with the corresponding untreated plants. The ionic fraction also decreased, but insignificantly (Table 1). In *Gossypium* plants untreated with kinetin the osmotic pressure of the leaf sap and its ionic fraction significant at -7 bar only (Table 2.) Kinetin treatments seem to reduce the total osmotic pressure insignificantly both in stressed and unstressed plants a compared with untreated plants. The ionic fraction in these plants exhibited the same trend, but the decrease was significant at -7 bar only. The analysis of variance of the data of osmotic pressure and its ionic fraction of the two experimental plants indicated that the F values for all factors were highly significant except in the case of the effect of kinetin on the total osmotic pressure in *Sorghum* plants. It should be recalled here that the treatment with kinetin by presoaking method resulted in a highly significant interaction between kinetin and salinity and high  $\eta^2$  values as compared with other methods with respect to the osmotic pressure either total or partial ionic.

The data of carbohydrate contents in both *Sorghum* and *Gossypium* plants either presoaked or not in kinetin solution are shown in Tables 1 and 2, respectively. Obviously the carbohydrate content (soluble and reserve) in *Sorghum* plants is higher (more than double) than that in the *Gossypium* plants. The soluble sugars in both experimental plants significantly decreased with increasing soil salinity as compared with the control plants. Meanwhile, the reserve sugars increased significantly in the same plants under the same conditions. Treatment with kinetin solution resulted in a significant increase in soluble sugars in both experimental plants as compared with the untreated salinized levels. The reverse occurred with respect to reserve sugars where the content decreased significantly in the treated plants of both experimental plants as compared with the corresponding untreated and salinized plants. It should be noted that, in the non stressed plants for the two plants tested, the carbohydrate content, either soluble or reserve, was not affected by the presoaking in kinetin solution. The statistical analysis of these data (Table 3)



indicates that F values for all factors were highly significant in both experimental plants except the F value of the effect of kinetin by the presoaking method on reserve sugars.

### Discussion

Retention of relatively high leaf water potential is believed to be essential for a normal metabolism in the plant. Osmotic adjustment to external salinity is not necessarily the result of excessive salt entry into the plant. As observed in the plants under investigation, the ionic partial osmotic pressure did not change appreciably with increasing soil osmolality, and significant increases in total osmotic pressure were found to be largely due to non-ionic (metabolic) material. Data also revealed that the osmotic potentials of the leaf sap in the two experimental plants increased with increasing soil salinity. Similar results were also obtained by some other authors (Bernstein 1961; Abdel-Rahman 1966; Abdel-Rahman et al. 1972 and Salama et al. 1980). In the present study, it was found that kinetin increased the osmotic pressure in salinity-stressed experimental plants, when applied by spraying and by irrigating water. However presoaking of seeds in kinetin solution resulted in a considerable reduction of osmotic pressure. This increase in osmotic pressure may be due to an increase in the soluble metabolic material. This agrees with the obtained results, that kinetin treatment increased the soluble carbohydrate contents significantly in stressed plants, which indicates that the increase in osmotic potential was achieved by increasing soluble sugars in the two experimental plants. It was also found that the increase in soluble carbohydrate occurred at the expense of the reserve sugars, which indicates that kinetin may cause the conversion of the insoluble carbohydrate (reserve) into soluble sugars to increase the osmotic pressure. This increase in soluble sugars and decrease in the insoluble carbohydrates as a response to kinetin treatment was reported by Boothby and Wright 1962; Dennis et al. 1967; Street et al. 1970 and Berridge and Ralph 1971. Alternatively, the presoaking of seeds in kinetin resulted in relatively lower osmotic pressure than that in untreated plants despite the accumulation of soluble carbohydrate. This is mainly due to the increase in relative water content in these plants under the same conditions as indicated in a previous study (Salama and Awadalla 1986). This reduction in osmotic pressure after seed presoaking indicates that kinetin interacts with salinity only when seeds of the two plants were presoaked in kinetin solution.

### References

- Abdel-Rahman, A. A. (1966): Salt accumulation and its effect on the osmotic pressure of plants. *Bull. Fac. Sci. Cairo University*. **40**, 73-86.  
Abdel-Rahman, A. A., El-Shourbagy, M. N., Shalaby, A. F., El-Monayeri, M. O. (1972):



- Salinity effects on growth and water relations of some desert range plants. *Flora*, **161**, 495-508.
- Bernstein, L. (1961): Osmotic adjustment of plants to saline media I. Steady state *Amer. J. Bot.* **50**, 360-370.
- Berridge, M. V., Ralph, R. K. (1971): Kinetin and carbohydrate metabolism in chinese cabbage. *Plant Physiol.* **47**, 562-567.
- Black, C. A., Evans, D. D., White, J. L., Enaminger, L. E., Clark, F. E. (1965): *Methods of soil analysis*. No. 9. in the series "Agronomy" American Society of Agronomy. Madison, Wisconsin.
- Boothby, D., Wright, S. T. C. (1962): Effect of kinetin and other plant growth regulators on starch degradation. *Nature*, **196**, 389-390.
- Cooper, A. W., Dumbroff, E. B. (1973): Plant adjustment to osmotic stress in balanced mineral nutrient media. *Can. J. Bot.* **51**, 763-773.
- Dennis, D. T., Stubbs, M., Coulate, T. P. (1967): The inhibition of Brussels sprout leaf senescence by kinins. *Can. J. Bot.* **45**, 1019-1024.
- Dubois, M., Gilles, K. A., Hamilton, R. A., Rabers, R. A., Smith, F. (1956): Colorimetric method for the determination of sugars and related substances. *Ann. Chem.* **28**, 350-356.
- El-Sharkawi, H. M., Abdel-Rahman, A. A. (1974): Response of olive and almond orchards to partial irrigation under dry farming practices in semi-arid region. II Plant soil water relations in olive during the growing season. *Plant and Soil* (The Netherlands). **31**, 13-23.
- El-Sharkawi, H. M., Salama, F. M. (1973): Drought resistance criteria in some wheat and barley varieties. II. Adjustment in internal water balance. *7th Arab Sc. Congre. Cairo*. **5**, 25-42.
- El-Sharkawi, H. M., Salama, F. M. (1976): Salt tolerance criteria in some wheat and barley cultivars. II. Adjustment in internal water balance. *Bull. Fac. Sci. Assiut Univ.* **5**, 1-15, 1976.
- Hoagland, D. R., Arnon, D. I. (1950): The water culture method for growing plants without soil. *Calif. Agric. Exp. Sta. Cir.* 347-352.
- Lagerwerff, J. V., Eagle, H. E. (1961): Osmotic and specific effects of excess salts on beans. *Plant. Physiol.* **36**, 472-477.
- Mothes, K. (1964): *The role of kinetin in plant regulation*. (In: *Regulateurs Naturels de la Croissance vegetale*.) Center National de la Recherche Scientifique, Gif-sur-Yvette, France 131-140.
- Nevin, D. J., English, P. D., Albersheim, P. (1966): The specific nature of plant cell wall polysaccharides. *Plant. Physiol.* **42**, 900-906.
- Oertli, J. J. (1976): The physiology of salt injury in plant production. *Z. Pflanzenern. Bodenkde.*, **2**, 195-208.
- Ostel, B. (1963): *Statistics in Research*: The Iowa State University Press. Amer. Iowa, USA
- Pallas, J. E., Box, J. E. (1970): Explanation for the stomatal response of excised leaves to kinetin. *Nature*. **227**, 87-88.
- Parker, J. (1969): Further studies of drought resistance in woody plants. *Bot. Rev.* **35**, 317-371.
- Ploxinski, N. A. (1969): *Rucovedstvo po biometrii dlya zootechnikov*. Izdatelstvo "Kolos" Moskow
- Pucher, W. G., Leavensworth, C. S., Vickery, H. B. (1957): In: S. P. Colow and N. C. Kaplan (Eds). *Methods in Enzymology*. Acad. Press. New York.
- Salama, F. M., Khodary, S. E. A., Heikal, M. M. D.: (1980): Effect of saline irrigation and gibberelic acid on osmotic pressure, photosynthetic pigments and carbohydrate content of carrot and sugar beet plants. *Egypt. J. Bot.*, **23**, 113-121.
- Salama, F. M., Awadalla, A. A. (1986): Effect of kinetin and salinity on water relations of *Sorghum* and *Gossypium* plants. II. The relative water content. *Bull. Fac. Sci. Sohag, Assiut University*. In press.
- Simmelsgaard, S. E. (1976): Adaptation to water stress in wheat. *Physiol. Plant.* **37**, 167-174.
- Stocker, O. (1960): Physiological and morphological changes due to water deficiency. In: *Plant water relationships in arid and semi-arid conditions*. *Rev. of Research*, 63-104. UNESCO, Paris.
- Street, H. E., Simpkins, I. (1970): Studies on the growth in culture of plant cells. *J. Expre. Botany* **21**, 170-185.
- Tal, M., Imber, D. (1971): Abnormal stomatal behaviour and hormonal importance in Flacca, a wilt mutant of tomato. III Hormonal effects on the water balance in the plant. *Plant Physiol.* **47**, 849-850.
- Vaadia, Y., Raney, F. C., Hagan, R. M. (1961): Plant water deficits and physiological processes. *Ann. Rev. Plant Physiol.*, **12**, 265-292.



## INTERACTIVE EFFECTS OF WATER STRESS, TEMPERATURE AND $\text{NO}_3^-$ CONCENTRATION ON ALLOCATION OF SOLUBLE NITROGEN IN GERMINATING *BAUHINIA* SEEDS

H. M. EL-SHARKAWI and K. A. FARGHALI

BOTANY DEPARTMENT, FACULTY OF SCIENCE, ASSIUT UNIVESITY, ASSIUT, EGYPT

(Received, 11 may 1987; accepted 14 September 1987)

*Bauhinia* seeds were germinated for 20 days under 3-factorial treatment combinations of different levels of water potential,  $\text{NO}_3^-$  concentration and temperature in the incubation medium. Excised embryonic parts (radicle, plumule and cotyledons) were extracted with cold water and analysed for soluble protein and the content of total free amino acids. The effect of single factors and their interactions on the distribution of both nitrogen fractions in seed organs was variable, as was the reative role (share) of each in such effect. The data refer to a greater effect of temperature than water potential on nitrogen metabolism of *Bauhinia* seeds. As seeds of a leguminous species, those of *Bauhinia* apparently are not influenced by  $\text{NO}_3^-$  concentration in their incubation medium. The interaction between  $\text{NO}_3^-$  and either stress or temperature had also a minor effect on the allocation of both nitrogen fractions in germinating seeds organs.

**Keywords:** *Bauhinia* sp., factorial interactions, tropical adaptations, soluble proteins, total free amino acids

### Introduction

Multi-factorial laboratory experiments are necessary simulations of realistic natural field conditions. A careful choice of the main effective (intrinsic) factors in such experiments ensure a reasonable degree of compatible data obtained from field situations. In seed germination, three important soil factors interact: matric water potential ( $\psi_m$ ), temperature (T) and soil nitrogen ( $\text{NO}_3^-$ ). The effects of such factors, acting singly, are widely covered in the literature. Besides the single effect of each, four interactions may have a role in the germination processes ( $\psi_m \times T$ ,  $\psi_m \times \text{NO}_3^-$ ,  $T \times \text{NO}_3^-$  and  $\psi_m \times T \times \text{NO}_3^-$ ). According to Sharma Bain and Mercer 1966a, if significant interaction exists between two (or more) factors, the information obtained from studies on the single factor effect has only limited value. The knowledge of fundamental interactions of individual components of mineral nutrition and external environment is necessary for the solution of problems concerning the productivity of plants (Bain and Mercer 1966b).

Seeds of legumes, such as *Bauhinia*, usually contain large amounts of reserve materials in cellular organelles including protein bodies, lipid bodies



and starch grains in cotyledons that comprise the embryos. As germination proceeds, reserve materials in cotyledons are degraded by hydrolytic enzymes and utilized for the growth of the embryonic axis (Bewley and Black 1978). The germination process of some legumes is considered by several investigators to be divided into three or more phases (4.35) starting with breakdown of protein and starch reserves in the cotyledons and ending with growth of the embryonic axis as products of degradation are transported to the latter. The allocation of such products in the radicle, plumule and storage tissue after attainment of growth, therefore, should indicate the collective response of such processes to ambient external (intrinsic) factors.

The aim of the present work is to study the interactive effects of matrix stress (water potential,  $\psi_m$ ), temperature (T) and  $\text{NO}_3^-$  concentration on the allocation of both total free amino acids (A.A.) and soluble proteins (S.P.) in mature organs of germinating *Bauhinia* seeds. The relative roles of each single factor and their mutual interactions are assessed.

### Materials and methods

Solution of polyethylene glycol (PEG-4000, Union Carbide Corp., U.S.A.) in concentrations that give particular levels of water potential (0, -2, -5, -10 and -15 bar) were prepared. The merits of such solutions in simulating reduced matrix water potential ( $\psi_m$ ) are discussed elsewhere (Lawlor 1969; Lee and Takahashi 1966; Lowry, Rosenbrough, Farr and Bandall 1951; Metivier and Dale 1977). Other sets of PEG solutions at the same water potential were amended with nitrate (supplied as  $\text{KNO}_3$ ) in concentrations of 0, 250, 500, 700 and 1000 ppm for each  $\psi_m$  level. These were used in testing the combined effects of  $\psi_m$  and  $\text{NO}_3^-$  at particular (test) temperatures.

Homogeneous seeds of *Bauhinia variegata* L., pretreated with  $10^{-3}$  mercuric chloride solution, were thoroughly washed and placed on chemically pure filter paper in sterilised glass Petri dishes. Ten seeds were used in each dish, to which was added 30 ml of the test solution (PEG or PEG +  $\text{NO}_3^-$ ) which was found sufficient to support the seeds for the period of incubation, as the dishes remained covered. Three dishes (replicates) were assigned at random to each treatment level. The seeds were incubated in the dark at different constant temperatures (27 °C, 33 °C and 37 °C) in incubators with air circulation. The seeds failed to germinate at -15 bar under any of the test temperature.

After 20 days (at maximum germination percentage) both plumules and radicles as well as the remaining parts of seeds (storage tissue exclusive of the testa) in every Petri dish were rapidly washed in distilled water to remove the treatment solutions and dried between filter paper. The excised organs were then weighed and immediately blended in 10 ml of ice-cold distilled water and the supernatant was kept in deep freeze until the time of analysis.

In aliquot samples from extracts of different seed organs, total free amino acids (A.A.) and soluble proteins (S.P.) were determined according to procedures described by Lee and Takahashi (1966) and Lowry et al. (1951), respectively.

Statistical inferences necessary to evaluate the relative roles of single factors and their interactions in the different germination phases included the analyses of variance (F values) and coefficient of determination,  $\eta^2$  (12). The latter ( $\eta^2$ ) is a statistic used to evaluate the relative role (share) of each of the single factors, as well as their mutual interactions in contributing to the total effect of treatment (combinations), usually expressed as a percentage or a fraction (cf. 6).



## Results

### *Total content of soluble proteins (S.P.) and free amino acids (A.A.) in germinating seeds*

#### *Effect of stress*

The effects of matric stress and  $\text{NO}_3^-$  concentration at three different temperatures on total contents of S.P. and A.A. are illustrated in Fig. 1. Stress caused a general decrease in the total content of both nitrogen fractions at relatively low and relatively high temperatures (27 °C and 37 °C, respectively). At optimum temperature (33 °C) the response to stress was different. An increase in both S.P. and A.A. was observed at certain ranges of stress, apparently modified by  $\text{NO}_3^-$  level in the medium. Thus, total soluble proteins generally tended to increase at  $\psi_m$  below 0 bar and this increase continued at higher stress levels, the magnitude depending on the  $\text{NO}_3^-$  concentration in the medium. In the absence of  $\text{NO}_3^-$ , a decrease in S.P. was observed at  $\psi_m = 0$  to -2 bar. An increase was limited to the  $\psi_m$  range -2 to -10 bar; at -15 bar S.P. declined to almost nil. In the presence of  $\text{NO}_3^-$  at any concentration, an increase in S.P. content occurred as  $\psi_m$  decreased from 0 to -10 bar. This increase in S.P. was also observed with decreasing  $\psi_m$  from 0 to -2 bar at 37 °C. The response of total free A.A. to stress at optimum temperature (33 °C) is quite different from that of S.P. An increase in A.A. took place as  $\psi_m$  decreased from 0 to -10 bar, both in the absence of  $\text{NO}_3^-$  or at  $\text{NO}_3^-$  concentrations 250 to 750 ppm. At higher  $\text{NO}_3^-$  concentration (1000 ppm), the increase in A.A. was terminated at  $\psi_m = -5$  bar. Maximum A.A. content was recorded at  $\psi_m = -10$  bar or -5 bar; it decreased with further decreasing  $\psi_m$ . However, with  $\text{NO}_3^-$  concentrations 0 to 500 ppm a decrease in A.A. content was observed at  $\psi_m = -2$  to -5 bar. At relatively higher temperature (37 °C), A.A. content increased at  $\psi_m = -2$  to -5 bar with  $\text{NO}_3^-$  concentration of 0 to 750 ppm and at  $\psi_m = -2$  to -10 bar with  $\text{NO}_3^- = 1000$  ppm.

#### *Effect of temperature*

Both S.P. and A.A. contents were much higher at 33 °C than at either 27 °C or 37 °C, especially at intermediate  $\psi_m$  levels (-2 to -10 bar) and at ambient  $\text{NO}_3^-$  concentrations of 250 to 1000 ppm (Fig. 1). In the absence of  $\text{NO}_3^-$ , however, temperature had little or no effect on S.P. content, but the concentration of A.A. was much higher at 33 °C than at either the lower or higher temperature.



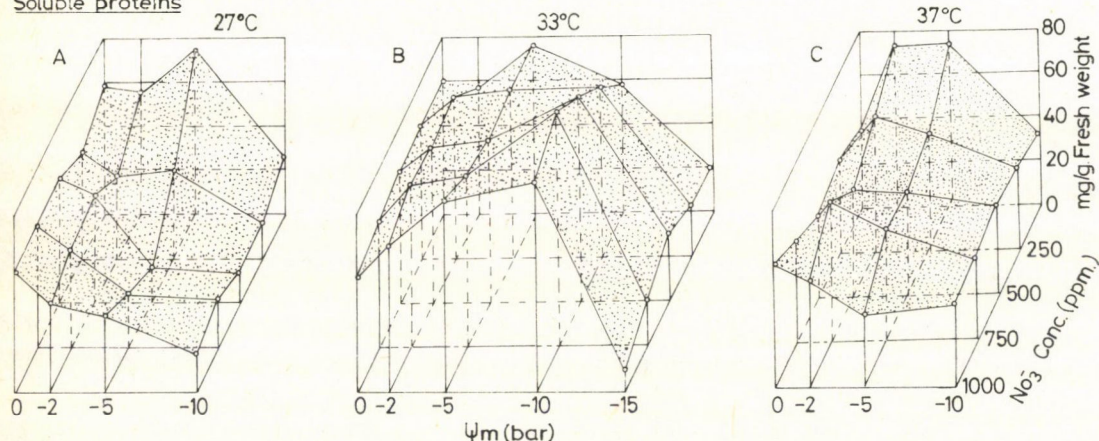
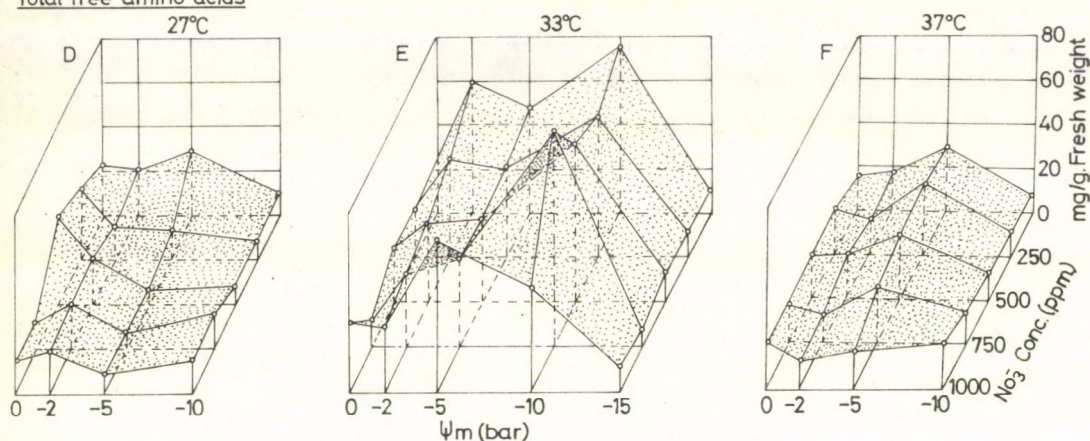
Soluble proteinsTotal free amino acids

Fig. 1. Total content of soluble proteins (A–C) and total free amino acids (D–F) in mg/gm F.wt. at different matric water potentials ( $\psi_m$ ), nitrate concentrations ( $\text{NO}_3^-$ ) and temperatures ( $T$  °C)

*Effect of nitrate*

Ambient nitrate concentration had no significant effect on either S.P. or A.A. content. In general, a slight decrease in both nitrogen fractions was observed with increasing  $\text{NO}_3^-$  concentration at nearly all stress levels. Under certain conditions an increase in S.P. content was observed with increased ambient  $\text{NO}_3^-$  concentration (at 33 °C and  $\psi_m = -10$  bar). Also, slight increases in A.A. content were observed with increased ambient  $\text{NO}_3^-$  concentration from 750 to 1000 ppm (at  $\psi_m = 10$  bar and 37 °C, and with  $\text{NO}_3^- = 511$  to 750 ppm at the same matric potential and 33 °C). The same was true with increased ambient  $\text{NO}_3^-$  concentration from 750 to 1000 ppm at -5 bar at the same temperature (33 °C).



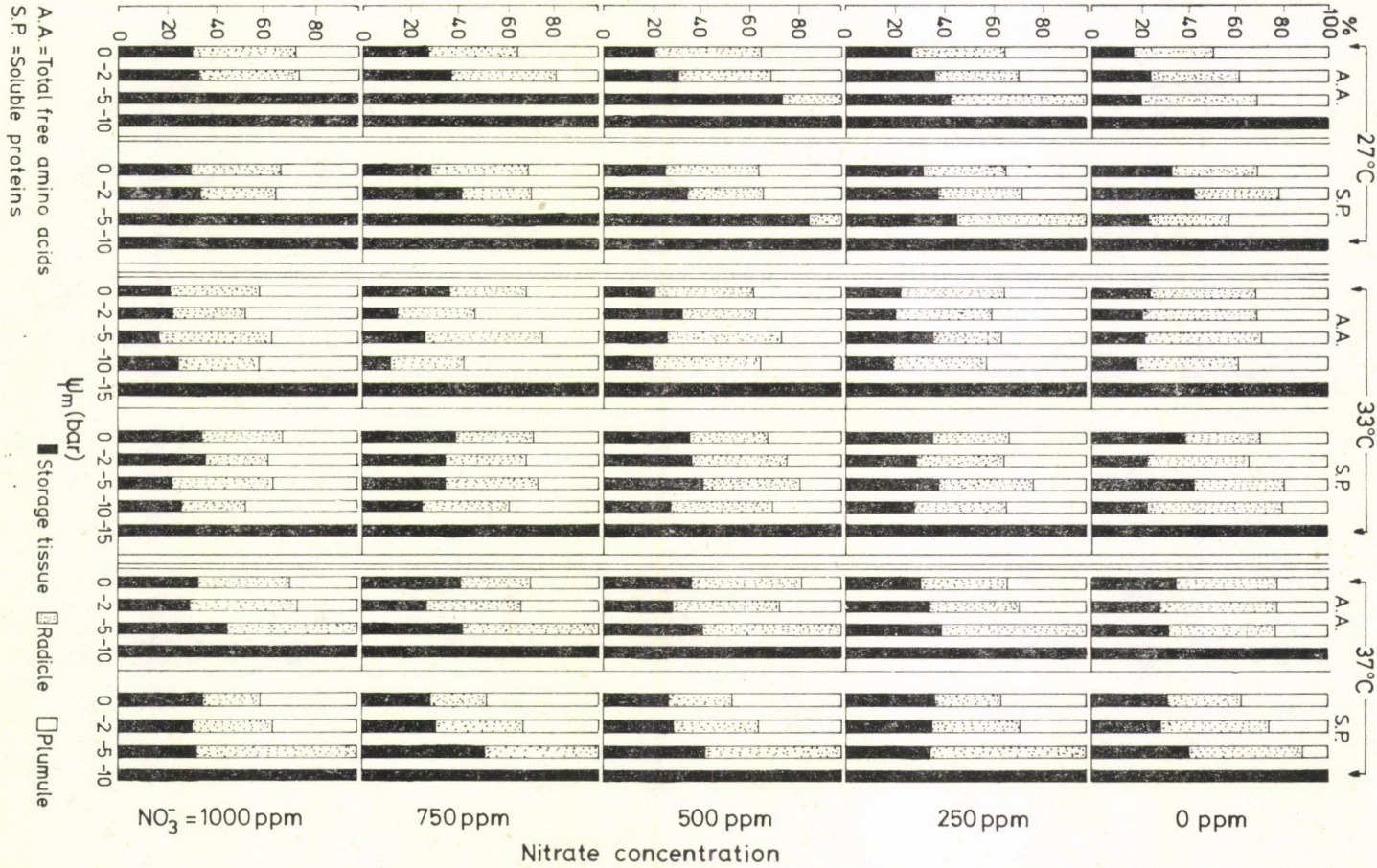


Fig. 2. Allocation of soluble proteins (S.P.) and total free amino acids (A.A.) in the radicle (R), plumule (P) and storage tissue (St.t.) at different matric water potentials ( $\psi_m$ ), nitrate concentrations ( $\text{NO}_3^-$ ) and temperatures ( $T^\circ\text{C}$ )

A.A. = Total free amino acids  
S.P. = Soluble proteins



*Allocation of free amino acids (A.A.) and soluble proteins (S.P.)  
in different seed organs*

The allocation of A.A. and S.P. in the embryonic axis and storage tissue at different levels of  $\psi_m$ ,  $\text{NO}_3^-$  at different temperatures is shown in Fig. 2.

The effect of temperature on the contents of both A.A. and S.P. in the plumule, radicle and storage tissue was highly significant (Table 1), similarly for the  $(T \times \psi_m)$  interaction. Matric water potential ( $\psi_m$ ) also had a highly significant effect on the content of both fractions in the storage tissue and on S.P. content in the plumule and the radicle. The role of temperature predominated among other single factors and interactions in their effects on the content of both A.A. and S.P. in the plumule, as indicated by  $\eta^2$  (Table 1). Meanwhile, the effect of  $(T \times \psi_m)$  interaction on both fractions was subdominant, as was the effect of  $\psi_m$  on S.P. In the radicle, the effect of T on A.A. content was predominant, whereas, the  $(T \times \psi_m)$  interaction was subdominant. The reverse

Table 1

*F and  $\eta^2$  values for the effect of T,  $\psi_m$ ,  $\text{NO}_3^-$  conc. and their interactions on the total free amino acids (A.A.) and soluble proteins (S.P.) in different organs of Bauhinia seeds*

Organ	Source of variance	(A. A.)		(S. P.)	
		F	$\eta^2$	F	$\eta^2$
Plumule	T	43.3**	0.48	17.4**	0.26
	$\psi_m$	2.5	0.04	9.1**	0.20
	$\text{NO}_3^-$	0.3	0.01	0.3	0.01
	$T \times \psi_m$	7.6**	0.26	5.3**	0.23
	$T \times \text{NO}_3^-$	0.6	0.03	1.2	0.07
	$\psi_m \times \text{NO}_3^-$	0.9	0.06	1.1	0.10
	$T \times \psi_m \times \text{NO}_3^-$	0.9	0.12	0.8	0.13
Radicle	T	30.6**	0.39	12.5**	0.22
	$\psi_m$	1.6	0.03	7.1**	0.19
	$\text{NO}_3^-$	1.8	0.05	2.2	0.08
	$T \times \psi_m$	7.9**	0.30	5.8**	0.30
	$T \times \text{NO}_3^-$	0.5	0.03	0.1	0.01
	$\psi_m \times \text{NO}_3^-$	0.7	0.05	0.5	0.05
	$T \times \psi_m \times \text{NO}_3^-$	1.0	0.15	0.7	0.15
Storage tissue	T	16.8**	0.25	13.0**	0.15
	$\psi_m$	8.5**	0.18	25.8**	0.48
	$\text{NO}_3^-$	0.8	0.02	0.7	0.01
	$T \times \psi_m$	7.5**	0.33	3.4**	0.12
	$T \times \text{NO}_3^-$	0.9	0.05	0.7	0.03
	$\psi_m \times \text{NO}_3^-$	0.5	0.04	0.4	0.03
	$T \times \psi_m \times \text{NO}_3^-$	0.7	0.13	1.3	0.18

\*\* Significant at  $P < 0.01\%$ .

Table 2

*F* and  $\eta^2$  values for the effect of ( $T \times \psi_m$ ) interaction on total free amino acids (A.A.) and soluble proteins (S.P.) in different organs of *Bauhinia* seeds at different concentrations of nitrate

Organ	NO <sup>-</sup> conc. ppm	(A. A.)		(S. P.)	
		F	$\eta^2$	F	$\eta^2$
Plumule	0	1.3	0.38	1.5	0.40
	250	8.1**	0.79	14.7**	0.87
	500	4.2**	0.66	4.8**	0.69
	750	3.4**	0.61	1.9	0.46
	1000	3.0*	0.58	2.2	0.51
Radicule	0	2.0	0.48	1.5	0.41
	250	4.5**	0.67	1.7	0.44
	500	3.1*	0.59	8.6**	0.80
	750	2.9*	0.53	3.8**	0.64
	1000	2.5*	0.55	1.6	0.42
Storage tissue	0	2.7*	0.55	4.3**	0.66
	250	3.0*	0.58	2.0	0.47
	500	2.21	0.50	2.7*	0.55
	750	2.7*	0.56	3.5**	0.62
	1000	1.8	0.46	2.9*	0.57

\* Significant at  $P < 0.05\%$ .

\*\* Significant at  $P < 0.01\%$ .

was true for its effect on S.P. In storage tissue the effect of ( $T \times \psi_m$ ) interaction on A.A. content was predominant while  $T$  and  $\psi_m$  effects were subdominant. Meanwhile, the effect of  $\psi_m$  on S.P. content was predominant and that of  $T$ , ( $T \times \psi_m$ ) and ( $T \times \psi_m \times \text{NO}_3^-$ ) was subdominant.

At no time did  $\text{NO}_3^-$  nor any interaction employing  $\text{NO}_3^-$  have a significant effect on either A.A. or S.P. content in different seed organs.  $\text{NO}_3^-$  as a single factor had a minor or negligible share ( $\eta^2 = 0.01-0.08$ ) in the total effect of treatment. The trifactorial interaction ( $T \times m \times \text{NO}_3^-$ ) contributed a slightly higher share in the total treatment effect ( $\eta^2 = 0.12-0.18$ ).

At relatively low temperature (27 °C) and high ambient  $\text{NO}_3^-$  concentration (750–1000 ppm) both A.A. and S.P. remained confined to the storage tissue at  $\psi_m$  lower than –2 bar. At the same temperature the radicle and plumule had a share of both A.A. and S.P., depending on  $\psi_m$  level. In this respect, the plumule was more sensitive to matric stress. At the higher temperature (37 °C) there was no inhibiting effect of high ambient  $\text{NO}_3^-$  concentrations on the allocation of both A.A. and S.P. into the embryonic axis. The plumule had a better chance in such allocation at 33 °C than at 37 °C, especially at moderate  $\psi_m$  (–5 bar).



*Effect of interactions**( $T \times \psi_m$ ) interaction*

The effect of ( $T \times \psi_m$ ) interaction at different  $\text{NO}_3^-$  levels is shown in Table 2. This interaction had a significant effect on total content of free amino acids in both the radicle and plumule at all  $\text{NO}_3^-$  concentrations. Its effect was not significant in the absence of  $\text{NO}_3^-$ . The relative role of this interaction tends to decrease with an increasing nitrogen concentration in both organs, as indicated by  $\eta^2$  values. Its effect on A.A. content in storage tissue was significant in the absence of  $\text{NO}_3^-$ , at 250 and 750 ppm.

This interaction apparently affects the content of soluble proteins in the seed organs in a different manner. In the plumule and radicle, no significant effect was found in the absence of  $\text{NO}_3^-$ . In the former, its effect was highly significant at relatively lower  $\text{NO}_3^-$  concentrations (250–500 ppm), whereas in the latter its effect was highly significant at higher  $\text{NO}_3^-$  concentrations (500–750 ppm). In storage tissue, the effect was significant either in the absence of  $\text{NO}_3^-$  or at relatively higher  $\text{NO}_3^-$  concentrations (500–100 ppm).

*( $T \times \text{NO}_3^-$ ) interaction*

Although the effect of this interaction on both nitrogen fractions was not significant. Total  $\psi_m$  levels (Table 1), it had a significant effect at particular  $\psi_m$  levels (Table 3). In the plumule it had a highly significant effect on

Table 3

*F and  $\eta^2$  values for the effect of ( $T \times \text{NO}_3^-$ ) interaction on the total free amino acids and soluble proteins in different organs of Bauhinia seeds at different matric potential levels*

Organ	$\psi_m$ bar	Total free amino acids		Soluble proteins	
		F	$\eta^2$	F	$\eta^2$
Plumula	0	14.3**	0.87	1.2	0.35
	- 2	3.2**	0.60	0.60	0.22
	- 5	3.3**	0.60	2.6*	0.55
	-10	0.6	0.20	0.5	0.17
Radicle	0	7.3**	0.77	4.3**	0.67
	- 2	1.8	0.46	1.1	0.34
	- 5	2.4*	0.53	1.29	0.38
	-10	0.3	0.11	0.2	0.07
Storage tissue	0	3.1**	0.59	3.7**	0.63
	- 2	0.8	0.28	0.7	0.23
	- 5	1.1	0.33	1.1	0.35
	-10	0.2	0.09	0.1	0.05

\* Significant at  $P < 0.05\%$ .

\*\* Significant at  $P < 0.01\%$ .

Table 4

*F* and  $\eta^2$  values for the effect of  $(\psi_m \times NO_3^-)$  interaction on the total free amino acids and soluble proteins in different organs of *Bauhinia* seeds at different temperatures

Organ	T °C	Total free amino acids		Soluble proteins	
		F	$\eta^2$	F	$\eta^2$
Plumule	27	9.5**	0.82	3.6**	0.62
	33	1.2	0.36	0.6	0.21
	37	6.8**	0.76	4.8**	0.69
Radicule	27	6.2**	0.74	2.6*	0.55
	33	1.0	0.32	0.4	0.14
	37	1.3	0.37	1.4	0.40
Storage tissue	27	2.2*	0.51	1.4	0.40
	33	0.6	0.22	0.5	0.21
	37	1.5	0.42	1.8	0.45

\* Significant at  $P < 0.05\%$ .

\*\* Significant at  $P < 0.01\%$ .

A.A. content over the  $\psi_m$  range 0 to -5 bar; but in storage tissue its effect was restricted to the level 0 bar. Its effect on S.P. content was significant only in the absence of matric stress ( $\psi_m = 0$  bar) in both the radicle and storage tissue and at  $\psi_m = -5$  bar in the plumule.

#### $(\psi_m \times NO_3^-)$ interaction

Despite its insignificant overall effect (over all temperatures), it yet has some significant effect at particular temperatures (Table 4). Its effect on both A.A. and S.P. content was highly significant in the plumule at 27 °C and 37 °C. In the radicle, the effect of  $(\psi_m \times NO_3^-)$  on the content of both nitrogen fractions was significant at 27 °C. The effect was significant only in the case of the A.A. content in the storage tissue at 27 °C.

#### Discussion

The data presented above clearly indicate that different seed organs varied in their response to the factors tested as well as to their interactions. Thus, temperature singly and its interaction with stress had a common significant role in both A.A. and S.P. content of different organs in germinating seeds of *Bauhinia*. However, the significant effect of stress ( $\psi_m$ ) on the content of both nitrogen fractions was limited to the storage tissue, and on S.P. content only in both the radicle and the plumule.

Nitrate interactions with either temperature ( $T \times NO_3^-$ ) or stress ( $\psi_m \times NO_3^-$ ) have no significant overall effect on the content of both nitrogen



fractions. However, both types of interaction did show some significant effect at particular levels of the third factors (stress and temperature, respectively). Thus the ( $T \times \text{NO}_3^-$ ) interaction showed a significant effect on A.A. content in the plumule at  $\psi_m = 0$  to  $-5$  bar, which was apparently hidden when its overall effect at different stress levels is considered. Likewise, it showed a significant effect in the absence of stress in both the storage tissue and the radicle at bar and in the radicle at  $-5$  bar. The effect of the same interaction on S.P. content was significant at  $\psi_m = 0$  bar in both storage tissue and radicle and at  $\psi_m = -5$  bar in the plumule. Also ( $\psi_m \times \text{NO}_3^-$ ) interaction had a significant effect at particular temperatures. Thus in the plumule, its effect on A.A. content was significant in all organs at relatively low ( $27^\circ\text{C}$ ) and relatively high ( $37^\circ\text{C}$ ) temperatures. Its effect on S.P. content was limited to the radicle and plumule at  $27^\circ\text{C}$  in the former and  $37^\circ\text{C}$  in the latter.

The fact that the matric potential had a significant effect on A.A. content only in the storage tissue (despite its significant effect on S.P. content in all organs of germinating seeds) may refer to the control of matric stress over translocation of A.A. from storage tissue into the embryonic axis. Even its relative role on S.P. content in storage tissue is much greater than any other factor or interaction (judged by  $\eta^2$  values, Table 1). As germination proceeds, reserve materials in the storage tissue (cotyledons) are degraded by hydrolytic enzymes and utilized for the growth of the embryonic axis (Bewley and Black 1978). According to Bain and Mercer (1966) the germination process in peas is divided into three distinct phases: (1) breakdown of protein and starch reserves during hydration of storage tissue, but such reserves are not lost from this tissue; (2) mobilization of hydrolysates into the embryonic axes; (3) utilization of such hydrolysates in the composition of the growing axes. Accordingly, no apparent effect of  $\psi_m$  is observed on the content of A.A. in the embryonic axis of *Bauhinia*, which signifies that the effect of matric stress on this nitrogen fraction is outside the embryonic axis, i.e. during its mobilization from the storage tissue. The data obtained, indicating that nitrate had no significant effect on the content of either A.A. or S.P. in all embryonic organs in *Bauhinia*, may be due to the fact that this plant and other legumes contain large amounts of reserve protein bodies which make the germinating seeds independent of  $\text{NO}_3^-$  concentration in the medium. The observed inhibitory effect of increased nitrate concentration on the allocation of A.A. and S.P. supports this view. Whether this has been through deactivation of enzymes involved, or feedback mechanisms, remains to be investigated otherwise, it appears that  $\text{NO}_3^-$  should be applied exogenously during germination or early seedling growth (Rodaway, Huany and Marcus 1979; Sharma 1973).

It was also observed that the interaction ( $T \times \psi_m$ ) always had a significant effect on both the A.A. and S.P. content in all seed organs and, in some instances, had a larger share in the total treatment effect than either T or



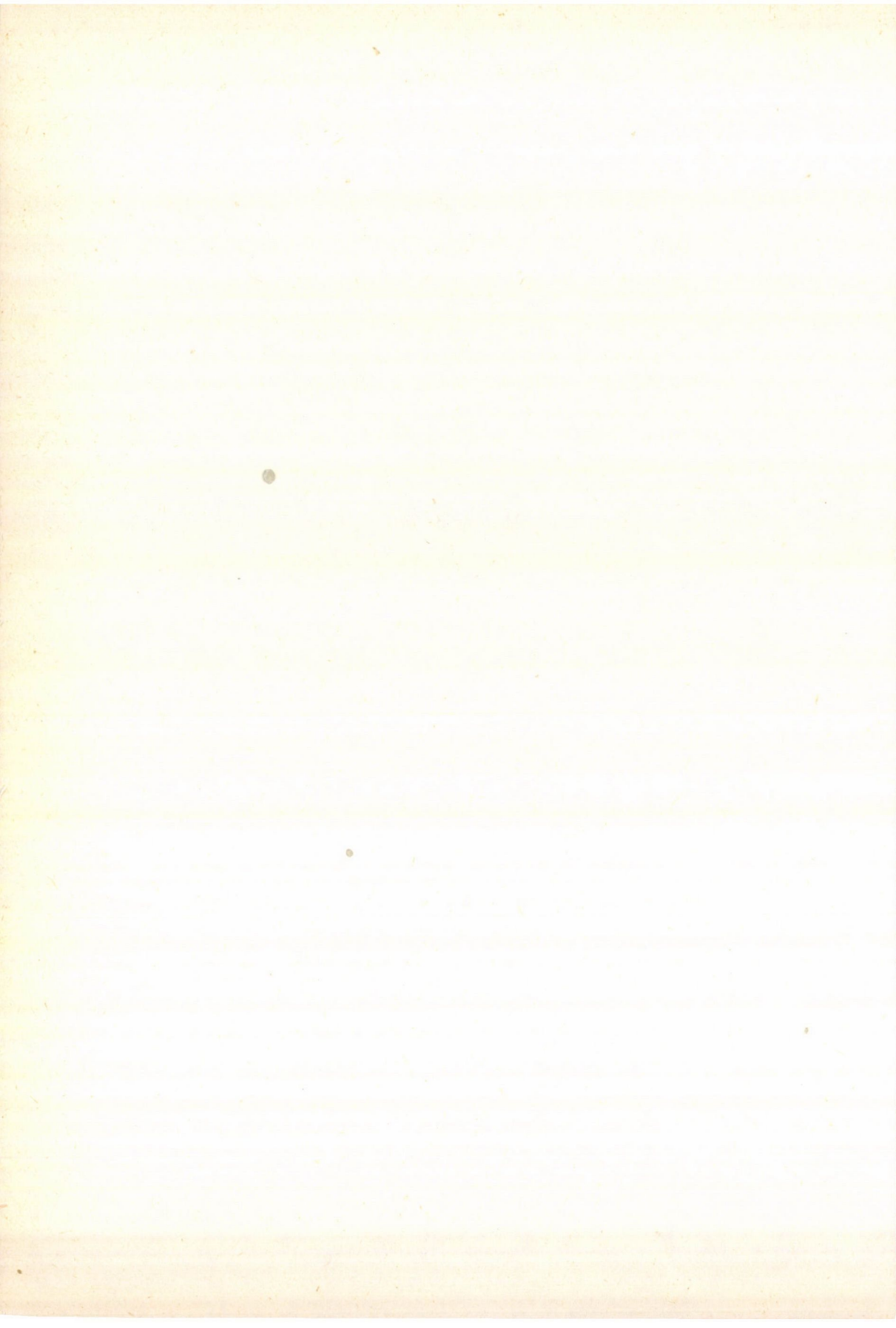
$\psi_m$  singly, as indicated by  $\eta^2$  values (for example, its effect on the S.P. of the radicle). The interaction of  $\text{NO}_3^-$  with either T or  $\psi_m$  had no significant effect on the contents of A.A. and S.P. in all organs of the germinating seeds. However, the trifactorial interaction ( $T \times \psi_m \times \text{NO}_3^-$ ) seems to have a relatively large share in the effect, though being non-significant.

Seeds of *Bauhinia*, apparently because of their tropical adaptations, are largely influenced in their nitrogen metabolism by temperature rather than by matrix stress. This has been previously observed by the authors (Sharma, 1976) in the germination of *Labonnychium* (a subtropical legume). In this regard these tropical legumes may be distinct from crop plants, germination of whose seeds is mainly influenced by  $\psi_m$  (Lawlor, 1969).

### References

- Bain, J. M., Mercer, F. V. (1966a): Sub-cellular organization of the cotyledons in germinating seeds and seedlings of *Pisum sativum* L. *Australian J. Biol. Sci.* **19**, 69–84.
- Bain, J. M., Mercer, F. V. (1966b): The relationship of the axis and the cotyledons in germinating seeds and seedlings of *Pisum sativum* L. *Australian J. Biol. Sci.* **19**, 85–96.
- Bewley, J. D., Black, M. (1978): *Mobilization of reserves*. In: Physiology and biochemistry seeds in relation to germination. Springer-Verlag Berlin, **1**, 177–244.
- El-Sharkawi, H. M., Springuel, I. V. (1977): Germination of some crop plant seeds under reduced water potential. *Seed Sci. and Technol.*, **5**, 677–688.
- El-Sharkawi, H. M., Farghali, K. A. (1985): Interactive effects of water potential and temperature in the germination of seeds of three desert perennials. *Seed Sci. and Technol.* **13**, 265–283.
- Lawlor, D. W. (1969): Plant growth in polyethylene glycol solutions in relation to the osmotic potential of the root medium and leaf water balance. *J., Exp. Bot.* **20**, 895–911.
- Lee, Y. P. and Takahashi, T. (1966): An improved colorimetric determination of amino acids with the use of ninhydrin. *Anal. Biochem.* **14**, 71–77.
- Lowry, C. H., Rosebrough, N. J., Farr, A. L., Bandall, H. J. (1951): Protein measurement with the folin phenol reagent. *J. Biol. Chem.* **193**, 265–275.
- Metivier, J. R., Dale, J. E. (1977): The utilization of endosperm reserves during early growth of barley cultivars and the effect of time of application of nitrogen. *Annal. Bot.* **41**, 715–728.
- Minamikawa, T., Suzuki, Y., Koshiha, T. (1983): Metabolic changes in axes of germinating *Vigna unguiculata* seeds as related to effects of removal of cotyledons. *Plant and Cell Physiol.* **24** (8), 1361–1369.
- Natr, L., Apel, P. (1983): The effect of phosphorus and nitrogen deficiency on growth of seedlings of spring barley in dependence on irradiance: growth analysis. *Biol. Plant.* **25**, 425–432.
- Natr, L., Apel, P., Fialova, S. (1983): The effect of phosphorus and nitrogen deficiency on growth of seedlings of spring barley in dependence on irradiance: content of chlorophyll, nitrogen and phosphorus. *Biol. Plant.* **25**, 433–439.
- Ostle, B. (1963): *Statistics in Research*. The Iowa State University Press Ames, Iowa, USA.
- Parmer, M. T., Moore, R. P. (1968): Carbowax-6000, mannitol and sodium chloride for stimulating drought conditions in germination studies of corn (*Zea mays* L.) of strong and weak vigor. *Agron. J.* **60**, 192–195.
- Rodaway, S. B., Huang, F., Marcus, A. (1979): *Nucleotide metabolism and germination of seed embryonic axes*. In the plant Seeds: Development, Preservation and Germination. (I. Rubenstein; R. L. Phillips, G. E. Green and B. C. Gengenbach, eds). Academic Press, Inc. New York, 203–218.
- Sharma, M. L. (1973): Simulation of drought and its effect on germination of five pasture species. *Agron. J.* **65**, 982–987.
- Sharma, M. L. (1976): Interaction of water potential and temperature effects on germination of three semiarid species. *Agron. J.* **68**, 390–394.





## Plant cultivation

# PHYTOMASS PRODUCTION OF MEDICINAL PLANTS IN FINLAND

B. GALAMBOSI

UNIVERSITY OF HELSINKI, DEPARTMENT OF HORTICULTURE\* FINLAND

(Received: 10 February 1987; accepted 24 June 1987)†

In 1984 and 1985, during 157 and 152 days of vegetation, 41 medicinal and herb plants were cultivated at 61st parallel of latitude in Finland.

The annual plants (*Silybum* sp., *Cnicus* sp., *Sinapsis* sp., *Brassica juncea*, *Borago* sp., *Oenothera* sp., *Calendula* sp., *Foeniculum* sp.) produced a phytomass of 33-65 t/ha in fresh and 8-10 t/ha dry form.

A phytomass of 20-33 t/ha fresh, and 4-7 t/ha dry weight was produced by *Anthriscus* sp., *Dracocephalum* sp., *Malva silvestris*, *Matricaria* sp., *Lepidium* sp., *Linum* sp., *Carum carvi* f. *annua*.

6-20 t/ha fresh and under 3 t/ha dry phytomass was produced by the heat-demanding plants *Majoranna* sp., *Ocimum* sp., *Thymus vulgaris*, *Carthamus* sp., *Saturea* sp., *Anethum* sp., *Artemisia annua*, *Coriandrum* sp.

The bi-annual and perennual plants generally produced 25-50 t/ha fresh and 5-8 t/ha dry phytomass in the second year. The fresh and dry root mass was 5-12 and 1-3 t/ha in the first year, 16-30 and 4-8 t/ha in the second year.

## Introduction

The value of the import of non-tropic origin herbs and medicinal plants to Finland in 1982 was 16.5 million FIM, including about 30 species (Hälvä 1985). In the fifties the possible cultivation of the most important spice and herb species has been studied, partly to reduce the spice import (Rautavaara 1953). During the eighties this question has arisen again associated with the study of the resources of the biologically active substances (essential oils, aroma compound, natural colour materials, bitter substances, etc.). The quantity of the theoretically potential production of herbs is about 860 tons, equal to 5 million FIM.

In 1984 the University of Helsinki established a new experimental station for studying the possibilities of cultivating aromatic and medical plants. Generally these plant species are known in Finland, and the main purpose of the experiments was to study the large scale cultivation possibilities among the given Finnish agricultural conditions.

\* The study was carried out in the "Puumalan Projekti" founded by research and Training Centre of the University of Helsinki.



**Table 1**  
*Meteorological data of the experiment*

Months	I.	II.	III.	IV.	V.	VI.
<b>1984</b>						
<i>Mean temperature</i>						
1930-60 (x)					8.6	13.9
1984 (xx)					13.6	14.0
Difference					+4.5	+0.1
<b>1984</b>						
<i>Rainfall</i>						
1930-60 (x)					40	57
1984 (xx)					22	72.3
Difference					-18	+15.3
<b>1985</b>						
<i>Mean temperature</i>						
1930-60 (x)	-9.1	-9.2	-5.3	1.8	8.6	13.9
1985 (xx)	-15.5	-18.5	-3.1	-0.2	10.1	13.2
Difference	+6.5	+9.5	+2.2	-2.0	+1.5	-0.7
<b>1985</b>						
<i>Rainfall</i>						
1930-60	42	30	28	33	40	57
1985	16.4	13.2	39.2	39.1	41.2	61.6
Difference	-15.6	-16.8	+11.2	+6.1	+1.2	+4.6

(x) = data of the South-Savo Exp. Station, Mikkeli

(xx) = data of the Puumala-Sorjola Meteorological Observation Station

The first step in the cultivation of plants in new ecological circumstances is to answer the biological questions. During 1984-1985 41 different plant species have been cultivated for the following information: what is the possible plant phytomass quantity under the determinate Finnish circumstances; what is the dynamics of plant development under the shorter vegetation period; what is the biological and technical maturity considering harvest possibilities, and what are the processing alternatives of the raw materials of natural origin?

This report contains the results of the cultivation of 25 annual and 16 biannual and perennial plants. Our aim is to answer the above questions.

### Materials and methods

#### *Climatic conditions*

The experimental station is located in Puumala commune at the centre of the lake Saimaa district at the northern latitude of 61°30' in Finland. The annual average temperature is 3.5 °C. The length of the growing period (V-IX) is 160-165 days the effective temperature

*Puumala (1984-85)*

VII.	VIII.	IX.	X.	XI.	XII.	V-IX.
16.7	14.6	9.4	3.6	-1.1	-5.4	12.6
15.8	14.0	9.2	5.5	1.0	-4.9	13.2
-0.9	+0.6	+0.2	+1.9	+2.1	-0.9	+0.6
69	73	61	61	48	43	300
112.8	54.5	123.2	103.8	36.8	37.2	384.8
+44	-18.5	+62	+42.8	-11.1	-5.8	+85
16.7	14.6	9.4	3.6	-1.1	-5.4	12.6
15.6	15.7	9.3	5.3	-3.0	-8.8	12.8
-1.1	+1.1	-0.1	+1.7	+1.9	+3.4	+0.2
69	73	61	61	48	43	300
128.2	119.6	78	73.6	61.5	74.4	428.6
+59.2	+46.6	+17	+13.6	+13.5	+31.4	+128.6

during the growing period is 1200-1300 °C. The annual precipitation is 560-670 mm, the precipitation of the growing period 300-325 mm.

The actual temperature and rainfall data are in the Table 1.

*Cultivation methods*

The type of the soil at the experimental station is a weakly-supplied stony morena soil. The results of the soil analysis are as follows:

Depth, cm	pH	Ca	K	P	Mg	N
				mg/l		
0-15	5.8	1100	55	9.6	45	14
15-30	5.9	1000	40	7.2	35	10

The quantity of fertilizer (10-16-17) was 500 kg/ha at the preparation of the soil in spring and 50 kg/ha N was spread twice.

The propagation material of plants originate mostly from the common varieties in Hungary, the others are given in Tables 2, 3 and 4. The cultivation and the observation have been carried out in 36 m<sup>2</sup> size plots.

The cultivation methods (space, seed-corn, care of plants) correspond to the general Hungarian practice (Hornok, 1978). In 1984 the direct sowing for seedlings in peat in the



plastichouse on april 25th, and the transplanting was on June 6th. In 1985 the corresponding dates were May 14–28, and June 19th. The harvest of annual plants generally took place between August 15 September–October 4. each year. In 1985, the two-year-old plants were cut first in July, secondly in September. At the marked phenological (Table 4.) phase  $2 \times 1$  m area was cut from the average of the large plots and the plants were dried to 8% of moisture content for phytomass calculation.

### Results and discussion

The results of the fresh and dry phytomass calculated for 1 hectare, are given in Tables 2, 3 and 4. The figures are the average of the two years, except for the *Cnicus* sp. and *Eruca* sp. which were cultivated only in 1984.

#### Annual plants

The length of the vegetation period was 152 and 157 days. It was sufficient for some of the studied species for developing a large quantity of green phytomass and reaching the biologically matured phase to produce germinative seeds. These species are the short breeding season plants: *Satureja* sp., *Dracocephalum* sp., *Anthriscus* sp., *Chamomilla* sp., *Linum* sp., *Borago* sp. and *Lepidium* sp.

Some of the longer breeding season plants — although they have produced a large quantity of green phytomass — since the biological maturity coincides with the technical maturity — could not give a utilizable product: *Foeniculum* sp., *Carum* sp., and *Oenothera* sp. The other part of the longer breeding season plants, though they could not give biologically mature seeds either, reached technical maturity and could be cut. These provided valuable products: *Origanum* sp., *Majorana* sp., *Ocimum* sp., *Anethum* sp., *Calendula* sp., *Cnicus* sp. *Malva* sp. and *Artemisia annua*.

According to the quantity of the phytomass, the plants can be divided into 3 groups:

- (1) Plants producing large quantity 33–65 t/ha fresh and 8–10 t/ha dry phytomass. The very tall plants belonging to this group are: *Sylibum* sp., *Cnicus* sp., *Sinapis* sp., *Brassica* sp. The high water containing leafy plants: *Borago* sp., *Oenothera* sp. and *Calendula* sp.
- (2) Low stature plants producing small quantity 5–10 t/ha fresh and under 3 t/ha dry phytomass. These are plants of mainly mediterranean origin, demanding warmth like *Origanum* sp., *Majorana* sp., *Satureja* sp., *Ocimum* sp., *Thymus* sp., *Salvia officinalis*, *Carthamus* sp., *Anethum* sp., *Artemisia annua*, *Coriandrum* sp. In this group the minimum factor was the heat quantity, for this reason *Ocimum* sp. was cultivated in a plastic house, an then the fresh phytomass increased from 5 t/ha to 18 t/ha.
- (3) Plants belonging to this group were generally 45–100 cm high and they developed 20–33 t/ha fresh and 4–7 t/ha dry phytomass. (*Dracocephalum* sp.,



**Table 2**  
*Phytomass production of annual herb plants*

Plant	Variety	Country	Height (cm)	Pheno-phase	Phytomass, t/ha		Drying rate
					fresh	dry	
<i>Anethum graveolens</i> L.	"Szilás"	Hungary	48.5	B	15.0	1.4	10.7
<i>Anthriscus cerefolium</i> (L.) Hoffm.	—	Hungary	59.5	E	33.7	3.3	10.2
<i>Artemisia annua</i> L.	—	Hungary	157.9	B	17.1	4.5	3.8
<i>Borago officinalis</i> L.	—	Hungary	90.6	D	46.7	5.3	8.8
<i>Brassica juncea</i> (L.) Czern.	"Trowse"	Hungary	144.5	E	37.1	9.9	3.7
<i>Calendula officinalis</i> L.	"Ball"	Hungary	77.7	D	41.0	5.3	7.7
<i>Carum carvi</i> L.	f. annua	Hungary	88.5	E	31.6	4.8	6.5
<i>Carthamus tinctorius</i> L.	"Budakalászi"	Hungary	70.3	D	17.8	5.2	3.4
<i>Chamomilla recutita</i> (L.) Rauschert.	"Budakalászi"	Hungary	61.7	D	25.4	4.5	5.6
<i>Cnicus benedictus</i> L.	—	Hungary	62.8	C	50.6	6.2	8.1
<i>Coriandrum sativum</i> L.	"Lucs"	Hungary	68.5	E	19.9	4.0	4.9
<i>Dracocephalum moldavica</i> L.	—	Romania*	55.1	D	23.3	4.4	5.2
<i>Eruca sativa</i> (Mill.) T.	—	Hungary*	120.7	E	32.2	10.0	3.2
<i>Foeniculum vulgare</i> Mill.	"Budakalászi"	Hungary	105.8	C	51.6	7.0	7.3
<i>Lepidium sativum</i> L.	—	Hungary	80.8	F	31.9	6.6	4.8
<i>Linum usitatissimum</i> L.	"Töp"	Finland	64.0	F	23.0	6.6	3.5
<i>Malva silvestris</i> L.	ssp. Maurit.	Hungary	147.5	D	31.9	5.9	5.4
<i>Ocimum basilicum</i> L.	"Keskenylevelű"	Hungary	27.2	D	4.9	0.7	7.0
<i>Oenothera biennis</i> L.	—	Hungary	91.5	E	49.7	7.7	6.4
<i>Origanum majorana</i> L.	"Francia"	Hungary	32.2	D	4.7	0.8	4.8
<i>Salvia officinalis</i> L.	—	Hungary	30.6	A	12.2	2.9	4.2
<i>Satureja officinalis</i> L.	"Budakalászi"	Hungary	31.2	D	20.7	4.1	5.0
<i>Sinapis alba</i> L.	"Gisilba"	Hungary	116.0	E	33.5	10.2	3.3
<i>Sylibum Marianum</i> (L.) Gaertn.	—	Hungary	174.4	E	63.5	14.7	4.3
<i>Thymus vulgaris</i> L.	—	Hungary	20.9	C	8.1	2.0	4.0

\* Origin of botanic garden.

*Marks of the phenophases:*

- A = sprouts without flowers
- B = start of budding
- C = start of flowering
- D = full flowering
- E = green seed phase
- F = start of seed ripening
- G = end of vegetation

*Anthriscus* sp., *Chamomilla* sp., *Linum* sp., *Lepidium* sp., *Malva* sp., etc.). The length of the vegetation period was a limiting factor from the standpoint of the number of harvest times. Contrary to the usual two harvest times in Central-Europe, in these experiments only one harvest could be done of



Table 3  
Phytomass production of biannual and perennial plants

Plant	Variety	Country	Age	Phenophase	Height (cm)	Phytomass, t/ha		Drying rate
						fresh	dry	
<i>Achillea collina</i> Becker	Cv. alba	Czechoslovakia	1 year	D	88.5	30.6	7.9	3.9
			2 year	D	83.1	32.9	8.1	4.1
<i>Artemisia absinthium</i> L.	—	Hungary	1 year	A	55.5	14.4	2.6	5.5
			2 year	C	145.5	33.6	8.4	4.0
<i>Barbarea vulgaris</i> R. Br.	—	Hungary*	2 year	E	84.0	30.6	8.7	3.5
<i>Hyssopus officinalis</i> L.	Blue-flower	Hungary	1 year	C	51.8	14.3	2.9	4.9
			2 year	D	50.6	17.7	3.3	5.3
<i>Melissa officinalis</i> L.	—	Hungary	2 year	C	67.2	13.8**	2.2	6.3
<i>Mentha piperita</i> L.	Mitcham	Hungary	1 year	B	50.0	18.1**	3.4	5.3
			2 year	B	70.0	24.4**	4.7	5.2
<i>Nepeta cataria</i> L.	—	Soviet Union*	2 year	C	95.0	38.5**	7.2	5.3
<i>Origanum vulgare</i> L.	—	USA	1 year	B	36.1	6.9	2.1	3.3
			2 year	D	64.0	21.8	4.7	4.6
<i>Salvia sclarea</i> L.	Akali	Hungary	2 year	D	117.0	51.1	7.9	6.5
<i>Satureja montana</i> L.	—	Hungary*	2 year	C	33.0	13.5	2.7	5.0
<i>Thymus serpyllum</i> L.	—	Hungary	2 year	B	32.5	17.7**	3.9	4.5
<i>Urtica dioica</i> L.	—	Hungary	1 year	C	63.3	20.6**	4.1	5.0
			2 year	C	105.5	14.8**	2.9	5.1

\* Origin of botanical garden.

\*\* Total of two harvests in one year.

Marks of the phenophases:

A = sprouts without flowers

B = start of budding

C = start of flowering

D = full flowering

E = green seed phase

F = start of seed ripening

G = end of vegetation

Table 4  
Phytomass production of root plants

Plant	Variety	Country	Age year	Pheno- phase	Parts of plant	Height, cm	Phytomass, t/ha		Drying rate
							fresh	dry	
<i>Angelica archangelica</i> L.	Budakalászi	Hungary	1	A	leaves	89.3	15.2	2.1	7.2
					roots	36.7	4.9	1.3	3.8
			2	G	stalk	154.0	25.7	3.7	6.9
					roots	49.6	27.0	6.7	4.0
<i>Levisticum officinale</i> Koch	Budakalászi	Hungary	1	A	leaves	72.1	25.3	3.4	7.4
					roots	29.0	12.3	3.0	4.1
			2	G	stalk	160.0	32.7	4.9	6.7
					roots	42.7	26.4	6.6	4.0
<i>Rumex alpinus</i> L.	—	Hungary	2	G	roots	48.1	29.8	8.5	3.5
<i>Valeriana officinalis</i> L.	—	Germany FR	1	A	leaves	81.4	17.8	2.7	6.6
					roots	34.0	12.0	2.0	6.0
			2	G	stalk	170.0	34.9	5.8	6.0
					roots	59.0	16.6	2.8	5.9

Marks of the phenophases:

- A = sprouts without flowers
- B = start of budding
- C = start of flowering
- D = full flowering
- E = green seed phase
- F = start of seed ripening
- G = end of vegetation



*Majorana* sp., *Dracocephalum* sp., *Satureja* sp., *Ocimum* sp., *Thymus* sp., *Salvia officinalis*. The last two species are perennials, but the frost damage in their plantations was 100%. Safe cultivation of these plants could only be annual in the future.

At the same time, because of the regular precipitation during the summer, two harvests were gathered of *Chamomilla* sp., *Cnicus* sp., *Anthriscus* sp. Precipitation plays quite an important role in the autumn harvest time. Comparing the drying ratio of plants calculated in the experiments to the ratio of southern countries, the figures in Finland were higher. In the herbs the ratio was above 7 : 1, and was 10 : 1 at the highest.

### *Bi-annual and perennial plants*

During the extremely cold winter of 1984/85 there was 18% frost damage in *Satureja montana* and 33% in the directly sowed *Hyssopus* plantations. In the other plantations frost did not cause any damage, and was uncommon in *Melissa* sp., *Nepeta* sp., *Salvia sclarea* plots.

The harvesting of these plants has been done in the phase of technical maturity mainly in the flowering time. With the exception of *Melissa* sp. and *Artemisia absinthium*, all plants produced fertile seeds in the end of the second year. In the first year of the cultivation 1 harvest, and in the second year 2 harvests, were possible. The plants were generally 50–100 cm high and suitable for mechanization.

Already in the first year profitable raw material was available from *Hyssopus* sp., *Achillea* sp., *Mentha* sp. In the second year nearly all plants developed large quantities of phytomass 25–50 t/ha fresh and 5–8 t/ha dry.

Of the root herb plants, *Angelica* sp., *Levisticum* sp. and *Valeriana* sp. produced fertile seeds in the second years. With all but *Valeriana* sp. the ratio of the underground and overground parts of the plants changed in the second year to the benefit of roots. In the continuously wet soil *Valeriana* sp. produces large but very light root systems. In the second year the leaf and root production was bigger by 30–40% than in the first year. The root production in the first year was 5–10 t/ha fresh and 1–3 t/ha dry; in the second year 16–30 t/ha fresh and 4–8 t/ha dry.

### *Processing possibilities*

Because of precipitation as shown in Table 1, the water content of the raw materials was rather high during the harvesting times. This fact indicates the importance of quick and effective drying methods to preserve the quality of fresh raw materials. Producing good quality dried drugs (flos, folium, herba, radix) is a little more risky in Finland than in the chemical processing methods:



of distillation and extraction. Based on these experiments, it may be stated that there are some possibilities in this part of Finland to produce quite large quantities per hectare of raw materials containing biologically active substances.

As the figures on the essential oil content and the composition of the oils demonstrated — not mentioned in detail — the essential oil production of the studied species does not differ at this latitude essentially from that in the southern countries.

Naturally the results of the experiments give only theoretical possibilities of producing different products from the phytomass. The implementation of these possibilities with some of these plants requires more detailed investigations in agrotechnics and processing.

### Summary

In 1984 and 1985, during 157 and 152 days of vegetation, 41 medicinal and herb plants were cultivated to study the potential phytomass at 64° parallel of latitude in Finland.

From the annual plants a large quantity of phytomass was produced by *Silybum* sp., *Cnicus* sp., *Sinapis* sp., *Brassica juncea*, *Borago* sp., *Oenothera* sp., *Calendula* sp., *Foeniculum* 33–65 t/ha in fresh and 8–10 t/ha in dry form.

A middle quantity of phytomass (20–33 t/ha fresh and 4–7 t/ha dry) was produced by *Anthriscus* sp., *Dracocephalum* sp., *Malva silvestris*, *Matricaria* sp., *Lepidium* sp., *Linum* sp., *Carum carvi* f. *annua*.

A low quantity of phytomass: (6–20 t/ha fresh and under 3 t/ha dry) was produced by the warmth-demanding plants *Majorana* sp., *Ocimum* sp., *Thymus vulgaris*, *Varthamus* sp., *Satureja* sp., *Anethum* sp., *Artemisia annua*, *Coriandrum* sp.

The length of the vegetation period was insufficient for seed ripening of *Foeniculum* sp., *Oenothera* sp., *Carum carvi* f. *annua* and the seed quality depended on the type of the species of mustards, *Coriandrum* sp. and *Linum* sp.

From the perennial species *Salvia officinalis*, *Thymus vulgaris* and *Foeniculum* sp., were 100% damaged by the frost so their cultivation can only be annual.

The bi-annual and perennial plants generally produced 25–50 t/ha fresh and 5–8 t/ha dry phytomass in the second year. Except for *Artemisia absinthium* and *Melissa* sp., all produced fertile seeds. The fresh and dry root mass was 5–12 and 1–3 t/ha in the first year, 16–30 and 4–8 t/ha in the second year.

The larger scale cultivation of medicinal plants in Finland in the future will be affected by the shorter, colder and wetter vegetation period. This fact emphasizes the importance of the choice of shorter breeding season species, the use of effective drying systems and processing methods of the fresh raw materials.

### References

- Hälvä, S. (1985): Consumption and production of herbs in Finland. *J. of Agriculturale*. 57, 231–237.





## EFFECTS OF PLOT AREA AND *AZOLLA* INOCULA ON GROWTH AND NITROGEN YIELD OF *AZOLLA PINNATA* WHEN INTERCROPPED WITH RICE

D. P. SINGH and P. K. SINGH

LABORATORY OF BLUE-GREEN ALGAE, CENTRAL RICE RESEARCH INSTITUTE, CUTTACK,  
ORISSA, INDIA

(Received: 23 February 1987; accepted 24 June 1987)

The effects of varying plot area (46.8–280.5 m<sup>2</sup>) and *Azolla* inocula (0.5 and 1.0 t fresh weight/ha) were studied on growth, N yield of *Azolla pinnata* R.Br. and rice yield, when grown without application of phosphorus and pesticide, in two wet seasons. The increase in plot area from 93.5 to 187.0 m<sup>2</sup> caused significant reduction in biomass and N yield of *Azolla*, but further increase in area to 280.5 m<sup>2</sup> had little effect on its productivity and N gain during both seasons. Growth and N yield (N<sub>2</sub>-fixation) of *Azolla* were comparable in plots of 46.8 and 93.5 m<sup>2</sup>. Inoculum of 1.0 t/ha produced significantly more biomass and N yield of *Azolla* than the inoculum of 0.5 t/ha, irrespective of plot area.

Use of *Azolla* with 30 kg N/ha (all applied as basal) produced 12.7–26.6% more grain yield over nitrogenless treatment, as against 20.0–30.9% increase obtained with the split application of urea at 60 kg N/ha. Despite differences in *Azolla* biomass and N yield, plot area and *Azolla* inocula levels did not show significant difference in grain and straw yields of rice.

**Keywords:** *Azolla pinnata*, inoculation, field size, nitrogen yield, rice

### Introduction

*Azolla*, a genus of aquatic ferns, is distributed throughout the tropical, sub-tropical and temperate regions of the world. It is commonly found floating on the water surface in ponds, ditches, channels, and rice fields (Singh, 1977), and is known to fix atmospheric nitrogen in symbiotic association with a heterocystous blue-green algae, *Anabaena azollae* (Moore, 1969; Singh, 1979a; Peters et al., 1982; Watanabe, 1982). *A. pinnata* has been extensively used as a source of organic nitrogen for rice crops in China (Liu, 1979) and Vietnam (Dao and Tran, 1979). The positive response of this fern on the growth and yield of rice has been reported (Singh, 1979a, b, 1982a, b; Rains and Talley, 1979; Watanabe, 1982; Singh and Singh, 1986a).

The fern is either grown and incorporated into the soil before transplanting or grown as an intercrop with rice. *Azolla* plants often accumulate in the corners of the plots when wind and wave actions are high (Singh 1979b) during the rainy season. Therefore, division of the larger fields into smaller plots was suggested to overcome this problem (Singh 1982b). There are no



reports on the influence of plot area on the spread and growth of this fern in paddy fields. Thus, the effects of varying plot area and *Azolla* inocula levels on growth, N yield of *Azolla* and rice yield were assessed in this study.

### Materials and methods

The field experiment was conducted in a randomized block design with three replications at Central Rice Research Institute, Cuttack farm during the wet seasons of 1984 and 1985. The soil of experimental plot was sandy clay loam with pH 5.8, organic carbon 0.81%,

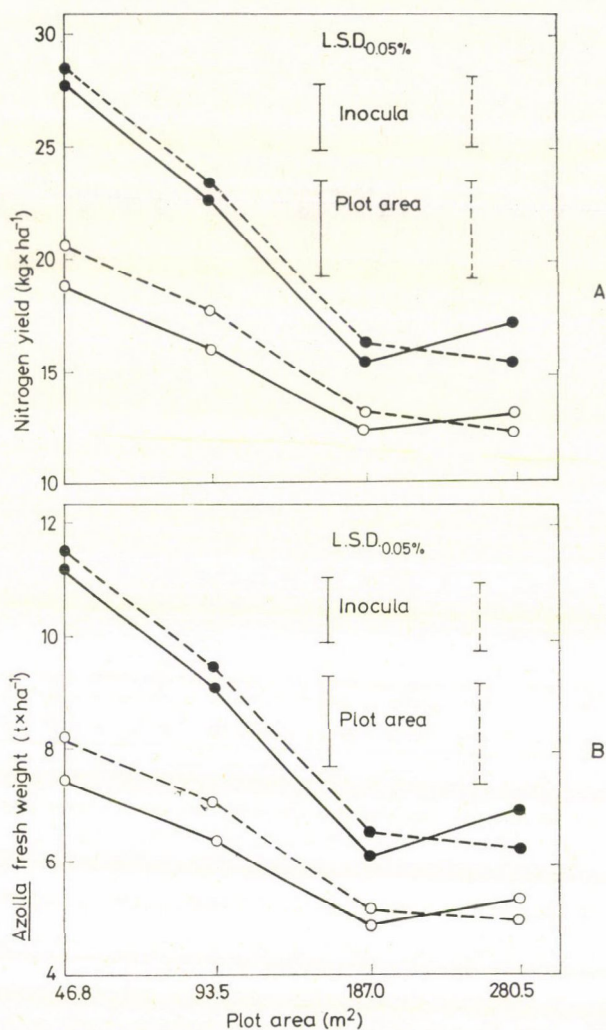


Fig. 1. Effect of plot area on nitrogen yield (A) and fresh weight (B) of *Azolla* intercropped at 0.5 (0) and 1.0 (0) t fresh weight/ha in wet seasons of 1984 (—) and 1985 (---)

total nitrogen 0.08% and available phosphorus 1.5 ppm (Olsen P). Rice seedlings (cv. IR 36) of 50 and 45 days age were transplanted in 1984 and 1985 respectively in the plots of 46.8, 93.5, 187.0 and 280.5 m<sup>2</sup> at a spacing of 20 × 15 cm. Fresh *Azolla pinnata* R. Brown (Thailand isolate) was inoculated at two rates of 0.5 and 1.0 t fresh weight/ha after 7 and 15 days of transplanting in 1984 and 1985 respectively. Urea at the rate of 30 kg N/ha was applied in these treatments before transplanting. Phosphorus and pesticide were not added to either *Azolla* or rice crop since their application increases the cost of *Azolla* production, and these materials are scare for many of the farmers. The plots of 46.8 m<sup>2</sup> were kept unfertilized while urea at the rate of 60 kg N/ha, half before planting, a quarter each at maximum tillering and panicle initiation stages, was applied in plots of 140 m<sup>2</sup> to serve as controls.

The growth (fresh weight t/ha) and N yield (kg N/ha) of *Azolla* were measured from 10, 20, 40 and 60 random samples respectively in plots of 46.8, 93.5, 187.0 and 280.5 m<sup>2</sup>. The samples were collected with the help of a 25 × 25 cm quadrat at 30 days of inoculation. Total nitrogen was estimated by the micro-Kjeldahl method (Jackson, 1967). *Azolla* was not incorporated into the soil and was allowed to decompose by itself. The data on grain and straw yields, number of panicles/m<sup>2</sup>, number of grains/panicle, thousand grain weight and sterility (%) were recorded at maturity of the rice crop. Treatments without nitrogen and 60 kg N/ha were not included in the statistical analysis, but were used for comparing the response of *Azolla* inoculation on the rice crop.

During the period of *Azolla* cultivation (September–October), rainfall, maximum/minimum temperature and relative humidity were 42–156 mm, 31.9–32.0/23.7–25.3 °C and 79–81% in 1984 and 289–343 mm, 30.5–31.0/22.4–25.3 °C and 80–88% in 1985 respectively.

## Results

### Growth and N yield of *Azolla*

*Azolla* grew well and covered all the plots in about 20–25 days of inoculation. The increase in plot area from 93.5 to 187.0 m<sup>2</sup> caused significant reduction in biomass and N yield of the fern during both wet seasons, but a further increase in the area to 280.5 m<sup>2</sup> had little effect (Fig. 1). The biomass and N yield of *Azolla* were comparable in the plots of 46.8 and 93.5 m<sup>2</sup>. *Azolla* applied

Table 1  
Effect of plot area and *Azolla* inocula on yield and yield components in rice

Plot area (m <sup>2</sup> )	Grain yield (t/ha)		Straw yield (t/ha)		Panicles/m <sup>2</sup>		Grains/Panicle		1000-grain wt. (g)		Sterility (%)	
	0.5	1.0	0.5	1.0	0.5	1.0	0.5	1.0	0.5	1.0	0.5	1.0
Wet season, 1984												
46.8	2.9	3.1	5.1	5.3	461	458	55	54	23	23	17	17
93.5	3.1	2.9	5.7	5.1	459	458	53	54	22	23	18	18
187.0	3.1	3.3	5.3	5.2	474	454	55	55	23	22	17	17
280.5	3.1	3.1	5.2	5.7	471	484	54	54	22	22	17	18
Wet season, 1985												
46.8	2.7	2.8	4.3	4.5	355	362	54	54	25	23	16	17
93.5	3.0	2.8	4.4	4.5	376	369	55	52	23	22	17	17
187.0	3.0	2.7	4.5	4.3	353	365	54	54	23	22	17	18
280.5	2.9	2.6	4.6	4.2	355	363	53	53	22	23	18	17

The treatment differences were statistically insignificant



at the rate of 1.0 t/ha produced 38.3% and 33.3% more biomass with 37.1% and 31.4% higher N yields than the inoculum of 0.5 t/ha in 1984 and 1985 respectively.

### *Rice yield*

The plot area and *Azolla* inocula levels had no significant effect on grain and straw yields, number of panicles/m<sup>2</sup>, number of grains/panicle, thousand grain weight and average sterility of rice in both wet seasons (Table 1).

*Azolla* intercropping with 30 kg N/ha (all as basal) produced 12.7–26.6% and 11.2–29.4% more grain and straw yields respectively over the nitrogenless control, whereas the application of 60 kg N/ha as urea increased these yields by 20.0–30.9% and 14.0–47.9% respectively. *Azolla* treatments were comparable to urea at 60 kg N/ha in increasing the grain number and in reducing the sterility of rice.

### Discussion

The studies on *Azolla* intercropping with rice were conducted mostly in smaller plots which might not give a precise estimate of its growth in relatively larger fields. In this study, we assessed growth and N yield of *Azolla* in plots up to an area of 280.5 m<sup>2</sup>, which approximate the field size of many Indian farmers.

Although *Azolla* can utilize combined nitrogen, it fixes atmospheric nitrogen efficiently when grown in rice fields. The nitrogenase activity of 99 nmoles ethylene produced (mg Chl.) min. was reported in fieldgrown *A. pinnata* (Manna and Singh, 1986), while it was 16 nmoles C<sub>2</sub>H<sub>4</sub> (mg Chl.) min. in mineral medium under net house conditions (Singh and Singh, 1986b). Thus, it fixed N<sub>2</sub> more efficiently when grown in flooded rice fields, either alone or in intercropping with rice.

Good growth of *Azolla* observed in this study without application of phosphorus was also reported by Watanabe and Ramirez (1984) in the Koronandal area of South Cotabato Province in the Philippines. The considerable *Azolla* biomass obtained in this experiment without addition of phosphate fertilizer was perhaps due to a relatively higher P content of the field soil, whereas the fern grew well without insecticide application because of fewer insects. Reduction in growth and N yield of *Azolla* with the increase in plot area might pertain to its accumulation at the sides due to wind and irrigation (Singh, 1979b). Singh (1982b) suggested the division of bigger fields into smaller plots to obtain good *Azolla* growth during this multiplication. Lumpkin and Plucknett (1982) suggested dividing larger fields, when it was to be produced in nurseries or even in intensive production of green manure. The



Vietnamese also recommended that rice fields be sub-divided into small squares to ensure control of the fern. The dikes were used to divide the larger fields into several subplots (Anonymous, 1982). The biomass and N yields of *Azolla* were higher in 1985 than in 1984, possibly because of more favourable environmental conditions during its cultivation. However, the reasons for higher *Azolla* fresh weight and N yield in 280.5 m<sup>2</sup> plots in 1984 are not clearly understood.

Growth and N yield of *Azolla* in 1.0 t/ha inoculum treatment were significantly more than those in 0.5 t/ha inoculum. Singh (1984) obtained 13% more biomass with inoculum of 1.0 t/ha than that used at 0.5 t/ha, when *Azolla* was intercropped with rice at 10 kg P<sub>2</sub>O<sub>5</sub>/ha. The relatively greater increase in *Azolla* growth at higher inoculum density in this trial could be due to the fern's growth in plots of varying area without phosphorus application. The positive response of inoculum density on growth and N<sub>2</sub>-fixation of *Azolla* was also reported in fallow fields (Dao and Tran, 1979; Sing, 1979a, 1982a).

The plot area and *Azolla* inocula levels did not differ in grain yield of rice, although growth and N yield of *Azolla* increased in smaller plots (upto 93.5 m<sup>2</sup>) and 1.0 t/ha inoculum treatment at 30 days. The reason for the insignificant difference in grain yield is not clear. However, it is likely that *Azolla* continued to grow in larger plots and thus contributed more N, whereas it decomposed early in smaller plots due to rapid mat formation. *Azolla* or urea-N also did not improve panicle production, perhaps because older rice seedlings of 45–50 days age were planted in the experiment. The increase in grain yield in *Azolla* treatments over the nitrogenless control was mainly due to a higher number of grains/panicle, probably because of reduced sterility. The positive response of *Azolla* on grain and straw yield, number of grains and sterility of rice crop has been reported (Singh, 1979a, b, 1982a, b; Talley and Rains, 1980; Watanabe, 1982; Singh and Singh, 1986a).

It is concluded from this study that *Azolla* intercropping without applications of phosphate fertilizer and pesticide, produced considerable biomass and N yield. Growth and N yield of *Azolla* at 30 days after inoculation in 93.5 m<sup>2</sup> plots were greater than those in 187.0 m<sup>2</sup> plots, while further increase in the area to 280.5 m<sup>2</sup> had little effect. The inoculum of 1.0 t/ha produced comparatively more biomass and N yield of *Azolla* than did the inoculum of 0.5 t/ha.

### Acknowledgements

Authors are grateful to Director, Central Rice Research Institute, Cuttack for providing necessary facilities.



## References

- Anonymous (1982): Report on the INSFER *Azolla* study tour in Vietnam. *Intl. Rice Res. Inst., Los Banos, Philippines*. 32-36.
- Dao, T. T., Tran Q. T. (1979): Use of *Azolla* in rice production in Vietnam. (In: Nitrogen and Rice.) *Intl. Rice Res. Inst., Los Banos, Philippines*. 395-405.
- Jackson, M. L. (1967): *Soil Chemical Analysis*. Prentice Hall of India, New Delhi, India.
- Liu, C. C. (1979): Use of *Azolla* in rice production in China. (In: Nitrogen and Rice.) *Intl. Rice Res. Inst., Los Banos, Philippines*. 375-394.
- Lumpkin, T. A., Plucknett D. L. (1982): *Azolla as a green manure*. Westview Press, Inc., USA.
- Manna, A. B., Singh P. K. (1986): Nitrogen fixation of *Azolla pinnata* (Bangkok) in presence of combined nitrogen sources in rice field. (In: R. Singh, H. S. Nainawatee and S. K. Sawhney, eds. Current Status of Biological Nitrogen Fixation Research.) *Haryana Agril. Univ., Hisar, India*. 209-210.
- Moore, A. W. (1969): *Azolla*: biology and agronomic significance. *Bot. Rev.* **35**, 17-34.
- Peters, G. A., Calvert, H. E., Kaplan, D., Ito, O. and R. E. Toia Jr. (1982): The *Azolla-Anabaena* symbiosis: morphology, physiology and use. *Isr. J. Bot.* **31**, 305-323.
- Rains, D. W., Talley, S. N. (1979): Use of *Azolla* in North America. (In: Nitrogen and Rice.) *Intl. Rice Res. Inst., Los Banos, Philippines*. 419-431.
- Singh, D. P., Singh P. K. (1982a): Relative effects of *Azolla pinnata* and its combination with chemical nitrogen fertilizer on growth, yield and N uptake of rice. *J. Agril. Sci. Camb.* **106**, 107-112.
- Singh, D. P., Singh, P. K. (1986b): Growth and nitrogenase activity in *Azolla* species. *Indian J. Plant Physiol.* **29**, 99-103.
- Singh, P. K. (1977): Multiplication and utilization of fern *Azolla* containing nitrogen-fixing algal symbiote as a green manure in rice cultivation. *Riso* **25**, 125-137.
- Singh, P. K. (1979a): Use of *Azolla* in rice production in India. (In: Nitrogen and Rice.) *Intl. Rice Res. Inst., Los Banos, Philippines*. 407-418.
- Singh, P. K. (1979b): Symbiotic algal  $N_2$ -fixation and crop productivity. (In: C. P. Mallick, ed. *Ann. Rev. Plant Sci.*, Kalyani Publishers, New Delhi, India. 37-65.)
- Singh, P. K. (1982a): *Azolla* as an organic nitrogen fertilizer for medium and low land rice. (In: Rev. Soil Sci. Agril. Chem. Vol. II. Indian Agril. Res. Inst., New Delhi, India. 236-242.)
- Singh, P. K. (1982b): *Azolla* and blue-green algal biofertilizer technology for rice. *Indian Farming* **32**, 3-8 and 21-22.
- Singh, P. K. (1984): Biological (algal) nitrogen fixation. *Final Technical Report of a ICAR Project. Central Rice Res. Inst., Cuttack, India*. 17-18.
- Talley, S. N., Rains, D. W. (1980): *Azolla filiculoides* Lam. as a fallow season green manure for rice in temperate climate. *Agron. J.* **72**, 11-18.
- Watanabe, I. (1982): *Azolla-Anabaena* symbiosis — its physiology and use in tropical agriculture. (In: Y. R. Dommergues and H. G. Diem. eds. *Microbiol. Tropical soils and plant productivity*. Martinus Nijhoff (Dr. W. Junk Publishers, The Netherlands. 169-185.)
- Watanabe, I., Ramirez, C. M. (1984): Relationship between soil phosphorus availability and *Azolla* growth. *Soil Sci. Plant Nutr.* **30**, 595-598.



## *Plant genetics and breeding*

### HETEROSIS AND PATH COEFFICIENT ANALYSIS IN SESAME (*SESAMUM INDICUM* L.)

H. E. OSMAN

DEPARTMENT OF ARID LAND AGRICULTURE, KING ABDULAZIZ UNIVERSITY,  
JEDDAH — SAUDI ARABIA

(Received: 10, April 1986; accepted 15 December 1986)

Eleven male-sterile  $\times$  male-fertile hybrids and their eleven parents were evaluated for one season at the USDA Cotton Research Station in Shafter, California. Number of capsules per plant, capsule length, number of seeds per capsule, 100-seed weight, yield per plant, yield per hectare and oil content were determined.

The average yield per hectare of the hybrids was 51.3% above that of the fertile parents. Other traits with the exception of seed yield per plant exhibited insignificant heterotic effects.

The correlations between yield per hectare of the fertile parent and the yield of the  $F_1$  ( $r = 0.752^{**}$ ) generations indicated that superior fertile parents could have been selected phenotypically. Number of capsules per plant and of seeds per capsule in the  $F_1$ 's and of capsules per plant in the parents were the most important yield contributing characters.

**Keywords:** *Sesamum indicum* L., sesame, heterosis, yield analysis

#### Introduction

Heterosis in Sesame (*Sesamum indicum* L.), a predominately self-pollinated crops, has been demonstrated by various workers (e.g. Riccelli and Mazzani, 1964; Srivastava and Singh, 1968; Sarathe and Dabral, 1969, Murty, 1975; Dixit, 1976; Kotecha and Yermanos, 1978; and Tyagi and Singh, 1981). As in many other crops, the magnitude of this heterosis was related to the degree of genetic diversity in the parent generation. Riccelli and Mazzani (1964) noticed that heterosis in sesame was more conspicuous in hybrids of cultivars from distant localities. Similarly, Murty (1975), in India, reported that heterosis in Indian  $\times$  exotic crosses was higher than in Indian  $\times$  Indian and exotic  $\times$  exotic crosses.

In an  $8 \times 8$  diallel cross, Kotecha and Yermanos (1978) found that heterosis was expressed by longer and higher number of capsules and greater production of seed yield. According to these workers, heterosis in yield ranged from -28.8 to 237.8% over the better parent. The ranges being -28.0 to 122.6% for number of capsules and -25.7 to 10.6% for capsule length. Riccelli and



Mazzani (1964) reported an average heterosis of 66.2% for 510 hybrids selected from a series of diallel crosses involving 32 cultivars. Sarathe and Dabral (1969) reported a wide range of heterosis in yield, 14.05—199.5% above the midparents, in seven hybrids of sesame. Heterotic effects for 1000-seed weight (Srivastava and Singh, 1968, Sarathe and Dabral 1969), number of seeds per capsule (Sarathe and Dabral, 1969), oil content (Murty, 1975), plants height, number of capsules per plant and for number of branches per plant (Tyagi and Singh, 1981) were reported in the literature.

Simple correlations and/or path coefficients analysis for many of the yield contributing characters in sesame were carried out and reported by various workers (e. g. Khidir and Osman, 1970; Osman and Khidir, 1974; Dixit, 1975; Gupta and Gupta, 1977; Shukla, 1983, etc.). All of these studies indicated the importance of number of capsules per plants as a direct high yield contributing character.

A knowledge of inter-relationships among seed and capsule characteristics is necessary if selection for the simultaneous improvement of these traits is to be most effective. The study reported in this paper was designed to:

- (1) furnish information on the nature of these associations in male-sterile  $\times$  male-fertile hybrids and their parents.
- (2) measure the extent of heterosis in the  $F_1$ -hybrids and thereby establish the usefulness of the present male-sterile stock in hybrid sesame breeding programs.

### Materials and methods

Eleven inbred sesame lines selected from the University of California sesame breeding nursery at Riverside were crossed to a male-sterile stock (Ms) isolated from a tropical material obtained from D. B. Mazzani, Centro de Investigaciones, Agropecuarias, Maracay, Venezuela. Eleven  $F_1$  hybrids were obtained. Five of the parents (Morada, Maporal, Padella, A/1/2 and Tozi) were introduced from the tropics. These were true breeding lines; tall, heavily branching, late maturing and typical of tropical cultivars.

The other six lines were derivatives of crosses between temperate varieties. All six lines were nonbranching, medium-maturing and typical of temperate cultivars. As far as could be ascertained, the lines were not closely related. The origin and characteristics of each of the eleven lines are given in Table 1.

In the summer of 1979 a yield trial, which contained the eleven parents and their eleven hybrids, was planted at the USDA Cotton Research Station, Shafter, California. Each of the 22 entries was planted in a one row plot of  $1.0 \times 7.5$  m that was boarded by an  $F_2$ -segregating material. The experiment was replicated three times in a randomized block design.

Prior to harvest, 10 competitive plants were selected from the central 4.2 m of each plot and were used to determine number of capsules per plant, capsule length, number of seeds per capsule, 1000-seed weight, yield per plant and oil content. The yield per hectare was determined by seed yield harvested from the central area of 4.2 m<sup>2</sup> in each plot.

Heterosis was expressed as the percentage increase of the hybrid performance above the fertile parent. Differences in each parent — hybrid pair were statistically tested for significance and compared to those of the remaining pairs by using a *t*-test for paired comparisons. Correlation coefficients for all pairs of traits were calculated and some of them were analysed further by a path coefficient described by Dewey and Lu (1959). The correlations between generations were calculated for seed yield.



### Results and discussion

The averages for the parents<sup>3</sup>  $F_1$ 's and percent heterosis for the measured traits are given in Table 2. The parental lines ranged in yield from 1.1 to 11.8 g/plant (89 to 1048 kg/ha) and none of them had excelled the best hybrid  $M_s \times \times$  UCR 203. The  $F_1$ 's ranged from 3.4 to 16.4 g/plant (292 to 1502 kg/ha) with heterosis ranging from -19.6 to 540.0 percent (Table 2). Average heterosis was greatest and significant for seed yield per hectare ( $P = 0.01$ ), and yield per plant ( $P = 0.05$ ), and modest for number of seeds per capsule, number of capsule per plant and 1000-seed weight. Capsule length had a negative heterosis while oil content did not exhibit appreciable heterosis (Table 2).

Table 1

*Name, origin and characteristics of the parental lines as measured in the present study characters and code*

Line	Origin	No. of capsules/ plant	Capsule length mm	No. of seeds/ capsule	1000-seed wt. (g)	Yield/ plant (g)	Yield/ ha (kg)	Oil content (%)
		1	2	3	4	5	6	7
Lucidi	California	60	37	58	3.4	7.4	916	54.1
UCR2	California	70	44	54	3.5	6.9	906	55.0
UCR14	California	59	30	58	3.2	8.0	825	52.9
UCR23	California	97	32	58	3.0	11.5	849	51.0
UCR203	California	64	49	55	3.4	7.7	697	54.2
UCR234	California	95	49	53	3.5	11.8	1048	55.2
Morada	Venezuela	32	23	16	1.8	2.3	157	54.2
Maporal	Venezuela	28	21	9	2.3	0.8	89	53.7
Padella	India	31	28	12	3.7	1.6	157	53.0
A/1/2	Sudan	34	28	22	2.0	2.3	156	51.6
Tozi	Sudan	22	28	20	1.9	1.1	110	52.2

Table 2

*Average performance of parental and  $F_1$  generations and parent heterosis*

	Parents		$F_1$ 's		Percent heterosis	
	Mean	S. E.	Mean	S. E.	Range	Mean
1. No. of capsules/plants	53.8	6.5	60.2	8.8	-40-128	11.9
2. Capsule length (mm)	33.6	1.5	32.0	1.1	-24- 28	-4.9
3. No. of seeds/capsule	37.7	3.3	46.4	4.0	-34-255	23.1
4. 1000-seed wt. (g)	2.9	0.2	3.2	0.1	-10- 69	10.3
5. Yield/plant (g)	5.6	1.1	8.3	1.6	-35-361	48.2*
6. Yield ha (Kg)	522.6	116.3	790.8	123.7	-20-540	51.3**
7. Oil content (%)	53.4	0.4	54.1	0.5	-1- 7	1.3

\* and \*\* significant at 5% and 1% levels respectively



Although heterosis for yield and its components was striking in many cases, it is difficult to generalize as to the causes. Many workers, e.g. Riccelli and Mazzani (1964), Murty (1975) noticed that the magnitude of heterosis in sesame was directly related to both geographical and genetic diversity. In this study, however, the magnitude of percent heterosis was higher in the tropical hybrids ( $M_s \times$  tropical parents) than in the temperate hybrids ( $M_s \times$  temperate parents). Since both the male-sterile stock and the tropical parents were introduced from the tropics, it appeared that genetic diversity and not geographical diversity, was the key for this hybrid vigour. Geographical diversity need not be related to genetic diversity since varieties with the same geographic origin might have different genetic backgrounds and widely divergent features (Trehan et al. 1974).

Unfortunately, despite this high magnitude of percent heterosis, the tropical hybrids had significantly ( $P = 0.01$ ) lower yields than the temperate hybrids. This was to be expected, since the experiment was conducted in a temperate region. In this region the tropical cultivars tended to grow vegetatively, started fruiting very late in the season and consequently gave lower yields than the temperate cultivars (Table 1). It is questionable, however, whether the tropical hybrids will surpass the performance of their parents when grown in the tropics. If proved to be so this will probably stabilize and raise the sesame yields in areas characterized by their erratic and unpredictable rains.

Phenotypic correlation coefficients for all pairs of characters were calculated separately for the parents and the  $F_1$ 's. Yield per plant and yield per

Table 3

*Phenotypic correlations ( $D.F. = 9$ ) between traits in the parental and  $F_1$  generation*

No. of capsules/Plant (1)								
Capsule length (2)	P	0.718						
	$F_1$	0.517						
No. of seeds capsule (3)	P	0.855	0.755					
	$F_1$	0.613	0.824					
1000-seed wt. (4)	P	0.634	0.735	0.670				
	$F_1$	0.546	0.597	0.547				
Yield/Plant (5)	P	0.979	0.733	0.918	0.659			
	$F_1$	0.834	0.799	0.883	0.580			
Yield/ha. (6)	P	0.907	0.807	0.946	0.754	0.937		
	$F_1$	0.684	0.809	0.911	0.516	0.951		
Oil Content (7)	P	0.399	0.607	0.380	0.575	0.382	0.575	
	$F_1$	0.512	0.546	0.764	0.381	0.784	0.799	
		1	2	3	4	5	6	7

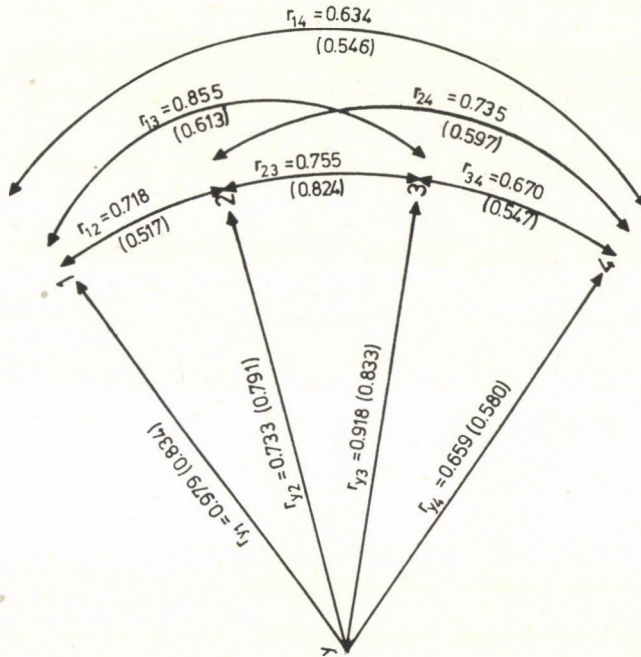


Fig. 1. Path diagram. Inter-relation between yield (y), number of capsules per plant (1), capsule length (2) number of seeds per capsule (3) and seed size (4) in 11F<sub>1</sub> hybrids (in parenthesis) and their parents

hectare were positively and significantly correlated ( $P = 0.01$ ) with each other and virtually with all the other characters in either the F<sub>1</sub>'s or the parental lines. Correlations of the yield contributing characters with each other and with oil content were positive. However, apart from those of number of seeds per capsule length, none of them was significant in both the F<sub>1</sub>'s and their parents.

A path coefficient analysis was used for further classification of the correlation coefficients. However, since the use of this method requires a cause and effect situation among the variables (Dewey and Lu, 1959), only four variables in addition to yield per plant were included in the analysis (Fig. 1). Yield per plant was considered the variable being affected, and number of capsules, capsule length, number of seeds per capsule and seed size (1000-seed weight) were considered the variables that affected it. The residual variable was assumed to be composed of all those factors that influenced yield per plant but independent of the remaining variables.

In both the F<sub>1</sub>'s and the parents, the number of capsules and number of seeds per capsule had the largest direct effect and indirect effects (through one another) on seed yield (Table 4). Direct effects of seed size were negligible



Table 4

*Path coefficient analysis of correlations between seed yield and four of its components in 11 F<sub>1</sub> hybrids and their parents*

Association and designation effect	Path coefficients	
	Parents	Hybrids
<i>Yield vs. No. of capsules per plant</i>		
Direct effect on seed yield	0.7294	0.4825
Indirect effect via:		
Capsule length	-0.0282	0.1178
No. of seed per capsule	0.2680	0.2632
Seed size	0.0100	-0.0297
Total correlation	0.9792**	0.8338**
<i>Yield vs. capsule length</i>		
Direct effect on seed yield	-0.0393	0.2277
Indirect effect via:		
No. of capsules per plant	0.5239	0.2496
No. of seeds per capsule	0.2366	0.3541
Seed size	0.0116	-0.0325
Total correlation	0.7328*	0.7989**
<i>Yield vs. No. of seeds per capsule</i>		
Direct effect on seed yield	0.3133	0.4296
Indirect effect via:		
No. of capsules per plant	0.5238	0.2957
Capsule length	-0.0297	0.1877
Seed size	0.0106	-0.0298
Total correlations	0.9180**	0.8831**
<i>Yield vs. seed size</i>		
Direct effect on seed yield	0.0157	-0.0544
Indirect effect via:		
No. of capsules per plant	0.4627	0.2633
Capsule length	-0.0288	0.1360
No. of seeds per capsule	0.2098	0.2351
Total correlations	0.6594*	0.5800
Residual effect (1-R <sup>2</sup> )	0.0166	0.0680

in both the F<sub>1</sub>'s and the parents, while these of capsule length were modest in the F<sub>1</sub>'s but relatively insignificant in the parental group.

The path analysis gave a somewhat different picture than did the simple correlation analysis. For instance, the analysis using the correlation coefficients as indices of effect gave the misleading impression that yield components,

Table 5

*Partial correlation coefficients for yield per plant (y), number of capsules (1), capsule length (2), number of seeds per capsule (3), and 1000 seed weight (4)*

Code***	Coefficient		Code***	Coefficient	
	Parents	Hybrids		Parents	Hybrids
$r_{y1.2}$	0.859**	0.586	$r_{12.3}$	0.213	0.018
$r_{y1.3}$	0.681*	0.443	$r_{12.4}$	0.480	0.223
$r_{y1.4}$	0.966**	0.626	$r_{13.2}$	0.686*	0.268
$r_{y2.1}$	0.211	0.535	$r_{13.4}$	0.750*	0.375
$r_{y2.3}$	0.082	0.128	$r_{14.2}$	0.186	0.286
$r_{y2.4}$	0.488	0.560	$r_{14.3}$	0.159	0.259
$r_{y3.1}$	0.202	0.563	$r_{23.1}$	0.391	0.617
$r_{y3.2}$	0.817**	0.384	$r_{23.4}$	0.503	0.607
$r_{y3.4}$	0.855**	0.685*	$r_{24.1}$	0.568	0.100
$r_{y4.1}$	0.242	0.518	$r_{24.3}$	0.471	0.212
$r_{y4.2}$	0.261	0.148	$r_{34.1}$	0.319	0.261
$r_{y4.3}$	0.149	0.143	$r_{34.2}$	0.259	0.082

\* and \*\* significant at 5% and 1% levels, respectively.

\*\*\*  $r_{y1.2}$  means variable y and 1 are correlated while variable 2 is kept constant . . . , etc.

number of capsules, number of seeds per capsule and capsule length had all more or less the same effect on yield in both the  $F_1$ 's and the parental lines; whereas the path analysis exposed capsule length as a minor influence, especially in the parental group (Table 4). The path analysis, unlike the simple correlation analysis indicated that some of the yield components, e.g., capsule length in the parents and seed size in the  $F_1$ 's had an opposing effect on the other characters. The apparent conflict between the results of the two analyses arises largely because the two methods measure different things. Whereas correlation simply identifies mutual association between the various parameters and varieties, the path analysis is concerned with the relative importance of each (Wright, 1921, Dewey and Lu, 1959). The path coefficient technique is, therefore, the more useful procedure when the goal is to establish direct and indirect inter-relationships among some of the variable as they affect yield components.

Correlation coefficients between generations were calculated for seed yield per hectare and seed yield per plant. The values between yields of fertile parents and  $F_1$ 's yields were 0.752\*\* and 0.596 for the respective characters, indicating that parental yield per unit area and not per plant was a good indicator of the  $F_1$ -performance.

The partial correlations recorded in this study (Table 5), similar to the path coefficients, emphasized the importance of number of capsules per plant and number of seeds per capsule for achieving high yields in breeding programmes.



The multiple correlation coefficients (R) computed for number of capsules per plant, number of seeds per capsule, capsule length and seed size were highly significant and their total contribution to yield was 97.9%\*\* in the parental generation and 86.9%\*\* in the hybrids.

### References

- Dewey, D. R., Lu, K. H. (1959): A correlation and path coefficient analysis of components of crested wheat-grass seed production. *Agron. J.* **51**, 515-518.
- Dixit, R. K. (1975): Path analysis for some quantitative traits in sesame (*S. orientale* L.) *Plant Science, India* **7**, 9-12.
- Dixit, R. K. (1975): Heterosis and inbreeding depression in sesame. *Ind. J. Agric. Sci.* **46**, 514-517.
- Gupta, V. K., Gupta, y. K. (1977): Variability, inter-relationships and path coefficient analysis of some quantitative characters in sesame. *Ind. J. Herd.* **9**, 31-38.
- Khidir, M. O., Osman, H. E. (1970): Correlation studies of some agronomic characters in sesame. *Expl. Agric.* **6**, 27-31.
- Kotecha, A., Yermanos, D. M. (1978): Combining ability of seed yield, plant height, capsule number and capsule length in an 8 × 8 diallel cross of sesame. (Abstracts). In *Agronomy Abstr.* Madison, U. S. A. *American Society of Agronomy*.
- Murty, D. S. (1975): Heterosis, Combining ability and reciprocal effects for agronomic and chemical characters in sesame. *Theor. Appl. Genet.* **45**, 249-299.
- Osman, H. E., Khidir, M. O. (1974): Relations of yield components in sesame. *Expl. Agric.* **10**, 97-103.
- Riccelli, M., Mazzani, B. (1964): Manifestations of heterosis in development, earliness and yield in diallel crosses of 32 sesame cultivars. *Agron. Trop. Venezuela*, **14**, 101-123.
- Sarathe, M. L., Dabral, K. C. (1969): Heterosis studies in *Sesamum orientale* L. *Sci. Cuh.* **35**, 572-573.
- Srivastava, D. P., Singh, S. N. (1968): Heterosis in sesame. *J. Ind. Bot. Soc.* **47**, 79-88.
- Shukla, g. P. (1983): Path coefficient analysis in sesame. *Ind. J. Agric. Sci.* **53**, 407-408.
- Trehan, K. B., Rao, A. V., Mehta, S. K., Chan, H., Sharma, H. N., Baijal, S. K. (1974): Genetic diversity in sesame. *Ind. J. Agric. Sci.* **44**, 208-212.
- Tyagi, B. P., Singh, H. G. (1981): Heterosis in sesame. *Ind. J. Agric. Sci.* **51**, 849-852.
- Wright, S. (1921): Correlation and causation. *J. Agric. Res.* **20**, 557-585.

## UTILIZATION OF CHLOROPHYLL AND LEAF MUTANTS IN F<sub>1</sub> HYBRIDS OF WATERMELONS AND MUSKMELONS

K. MOZSÁR, H. T. MINH and NADJA TAMÁSSY

PLANT GENETICS DEPARTMENT, UNIVERSITY OF HORTICULTURE AND FOOD INDUSTRY,  
BUDAPEST, HUNGARY

(Received: 12 February 1986; accepted 25 July 1987)

Treatments were carried out on a number of Hungarian and foreign watermelon and muskmelon varieties using chemical supermutagens. Stable xantha (xa) colour mutants were obtained in muskmelons and non-divided leaf (nl) mutants in watermelons. In recent years, partial diallel crosses have been carried out using these lines. Use was made of paternal testers which proved promising from the standpoint of earliness, yield ability, quality and disease resistance.

**Keywords:** Watermelon, muskmelon, chlorophyll-mutant, leaf-mutant

### Introduction

Throughout the world, more and more F<sub>1</sub> hybrid varieties are being utilized in the production of watermelons and muskmelons, since these have sufficient genetic plasticity to make good use of growing site conditions and thus have favourable production and marketing values.

The seed production of F<sub>1</sub> varieties is facilitated by the use of maternal lines bearing colour and leaf markers.

Marker genes for non-divided leaves are mentioned by Mohr et al. (1955), while Robinson et al. (1976) studied the inheritance of these genes. Investigations on this subject were made by Fehér (1985) jointly with the Plant Genetics Department. A colour marker for muskmelons is reported by Whitaker (1952). A description of the "halo" cotyledon marker in muskmelons is given by Nugent et al. (1974). The practical use of these markers in Hungary is reported by Velich (1985) and Muzik et al. (1985), who emphasize the decisive role of hybrid seed in quality.

### Materials and methods

For the crosses carried out in recent years the following mutant lines were used as the maternal partner:

"Xantha-1": this line has poor vegetative development, medium early ripening, medium yield potential, but extremely good quality. The fruit is oval, with a slight thin network pattern and green flesh. The leaves are bright yellow, acting as a good marker even in the cotyledon stage.



- "Xantha-74": this line has a medium vegetative mass, early ripening, medium yield potential, good quality and a pleasant aroma. The fruit is round, with a thin network pattern and yellow flesh. The leaves are pale yellow, but are suitable for use as a marker.
- "Non-divided leaf-28": an early line with good yield potential. The fruit is round, dark-green in colour and of medium quality. The first four foliar leaves are not divided, so provide a good marker.
- "Non-divided leaf-30": similar to the above in appearance, but the latest as regards ripening. Its quality and yield potential are medium. The first foliar leaves are only slightly divided, so can be used as a marker.
- "Non-divided leaf-46": an early line with intense vegetative development. The oval fruit are middle green with dark green stripes. It has high yield potential and medium quality. The leaves are non-divided up—to the fourth foliar leaf, so make a good marker. The muskmelon varieties "Magyarkincs", "Muskotály" and "Javitott-Zentai" and the watermelon varieties "Hevesi", "Sugar Baby" and "Smokylee" were used as paternal testers.

## Results

### *Evaluation of muskmelon $F_1$ combinations*

On the basis of the individual phenophases an examination was made of the vegetative and generative development and of the usefulness of the colour marker in the seedling stage.

The combinations listed in Table 1 are worth noting for their favourable vegetation periods. Particularly  $F_1$ s created using the "Javitott-Zentai" tester make early production possible.

**Table 1**  
*Phenophases of muskmelon  $F_1$  hybrids (1985)*

Combination	Development phases in days		
	Vegetative	Generative	Total
Xantha-1 $\times$ Magyarkincs	32.5	71.5	104
Xantha-1 $\times$ Javitott-Zentai	27.6	57.3	84.9
Xantha-74 $\times$ Magyarkincs	44.0	68.5	112.5
Xantha-1 $\times$ Javitott-Zentai	36.8	51.3	88.1

**Table 2**  
*Yields obtained for muskmelon  $F_1$  hybrids (1985)*

Combination	Mean mass of fruit, kg	No. of fruit/plant	Yield/plant, kg	%
Xantha-1 $\times$ "Magyarkincs"	0.78	3.6	2.52	119
Xantha-1 $\times$ "Javitott-Zentai"	1.18	4.7	5.53	176
Xantha-74 $\times$ "Magyarkincs"	0.98	2.9	2.65	101
Xantha-74 $\times$ "Javitott-Zentai"	0.92	2.8	2.45	107

**Table 3**  
*Quality of muskmelon  $F_1$  hybrids (1985)*

Combination	Dry matter	Sugar	Acid
	%		
Xantha-1 $\times$ "Magyarkincs"	9.01	8.39	0.34
Xantha-1 $\times$ "Javitott-Zentai"	5.96	6.13	0.32
Xantha-74 $\times$ "Magyarkincs"	11.36	10.70	0.55
Xantha-74 $\times$ "Javitott-Zentai"	10.21	9.57	0.46

Muskmelon colour markers enable the  $F_1$  percentage to be checked in the seedling stage, so hybrid seed can be bought up on a quality basis. When examining the yield of hybrid combinations, the seed setting (No. of fruit per plant) and the average mass of the fruit were used as the basis for differentiation between the performance of the combinations (Table 2).

The better parameters of the maternal line "Xantha-1" gave the most outstanding performance in combination with "Javitott-Zentai". The results achieved with Xantha-74 are less promising.

Among the quality indices, the soluble dry matter (Rf%), the percentage of inverted sugar and acid, which influence the flavour, were examined (Table 3).

On the basis of the quality indices, "Magyarkincs" proved to be the best partner, though the "Xantha-74"  $\times$  "Javitott-Zentai" combination is also of good quality.

#### *Evaluation of watermelon $F_1$ combinations*

As in the case of muskmelons, the evaluation was primarily aimed at determining the usefulness of maternal lines with non-divided leaves. Special attention was paid to the possible use of leaf markers in determining the  $F_1$  percentage (Table 4).

**Table 4**  
*Phenophases of watermelon  $F_1$  hybrids (1985)*

Combination	Developmental phases, days		
	Vegetative	Generative	Total
nl-28 $\times$ "Hevesi"	48	76	124
nl-30 $\times$ "Hevesi"	40	72	112
nl-46 $\times$ "Hevesi"	34	76	110
nl-28 $\times$ "Smokylee"	37	78	115
nl-30 $\times$ "Smokylee"	41	76	117
nl-46 $\times$ "Smokylee"	49	78	127

Note: nl = non-divided leaf mutant



**Table 5**  
*Yields obtained for watermelon F<sub>1</sub> hybrids (1985)*

Combination	No. of fruit/plant	Average mass/fruit, kg	Yield/plant, kg	%
nl-28 × "Hevesi"	1.78	2.60	4.68	92
nl-30 × "Hevesi"	1.42	3.00	4.26	107
nl-46 × "Hevesi"	1.42	2.10	3.88	97
nl-28 × "Smokylee"	1.64	3.80	4.24	102
nl-30 × "Smokylee"	2.02	2.55	5.16	128
nl-46 × "Smokylee"	2.02	3.28	4.75	116

Note: nl = non-divided leaf mutant

**Table 6**  
*Quality parameters for watermelon F<sub>1</sub> hybrids (1985)*

Combination	Dry matter	Sugar	Acid
	%		
nl-28 × "Hevesi"	8.34	9.85	0.19
nl-30 × "Hevesi"	8.12	10.40	0.34
nl-46 × "Hevesi"	9.18	11.70	0.41
nl-28 × "Smokylee"	7.95	9.36	0.28
nl-30 × "Smokylee"	8.39	9.74	0.52
nl-46 × "Smokylee"	9.13	10.20	0.47

Note: nl = non-divided leaf mutant

Among the non-divided leaf lines, numbers 30 and 46 gave better F<sub>1</sub>'s with both testers, though the vegetation period of "Hevesi" combinations was more acceptable than that of "Smokylee" combinations.

The late-maturing, high-yielding tester "Smokylee" increases the yield of the F<sub>1</sub> hybrids. Of the non-divided leaf maternal lines, nl-30 is the most favourable (Table 5).

In general, satisfactory values were obtained for the quality indices, particularly in the case of the tester "Hevesi" (Table 6). Among the non-divided leaf lines, the use of numbers 30 and 46 would again appear to be justified.

On the basis of examinations carried out in 1985 it can be stated that the available mutant muskmelon and watermelon lines carrying colour and leaf markers are suitable for the production of F<sub>1</sub> hybrids.

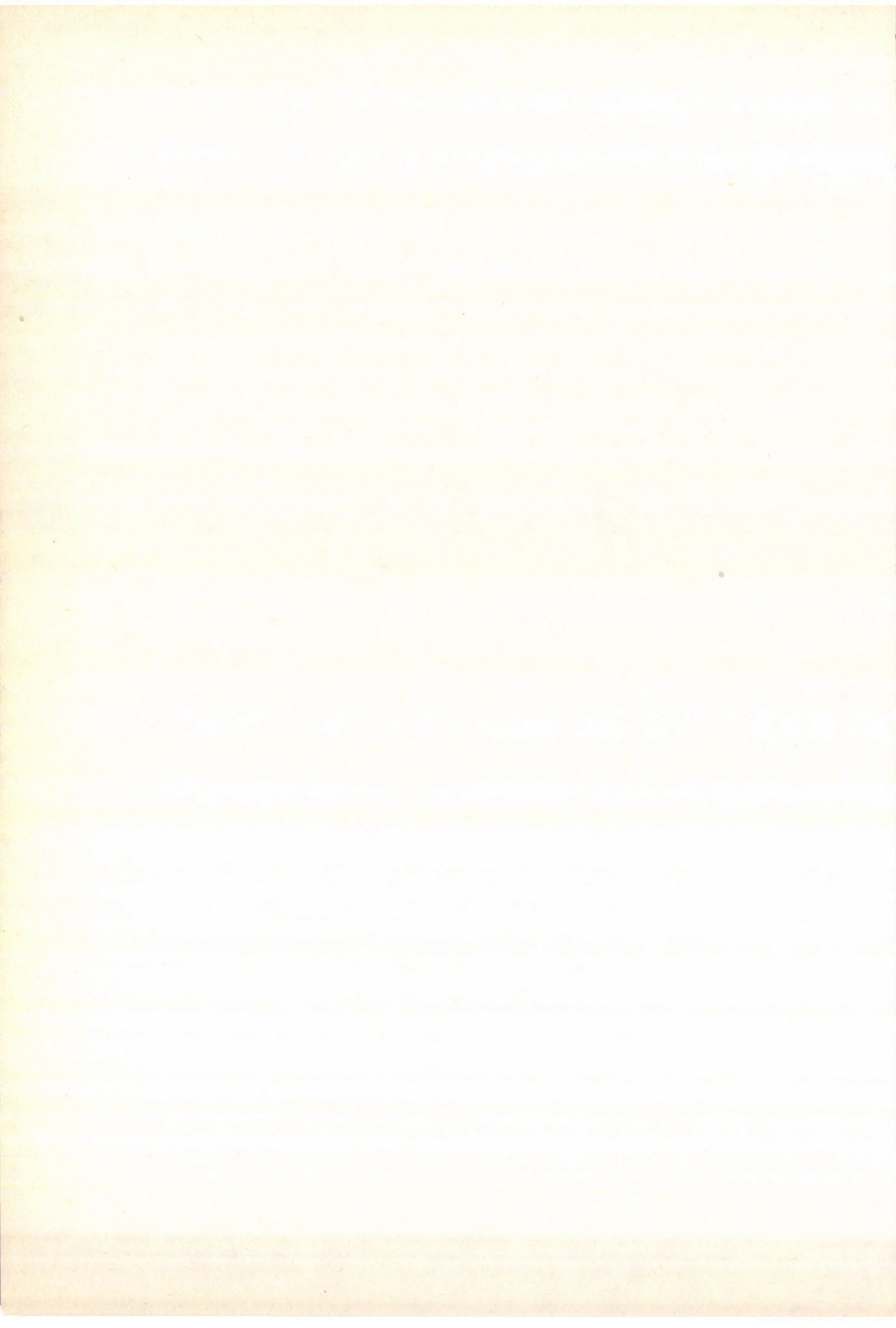
Light yellow foliar leaves (xantha) and non-divided leaves (nl) give an early indication of the hybrid percentage.

Further partial diallel crosses are planned using the available maternal lines bearing marker genes in order to produce  $F_1$  combinations with favourable production value for the selection of combinations with the best genetic potential.

### References

- Fehér, T. (1985): Use of markers with non-divided leaves in the breeding of watermelon hybrids. (A nem szeldelt levélmarker felhasználása a görögdinnye hibridnemesítésében.) *Zöldségtermesztési Kutató Intézet Bulletinje*, **18**, 5, 59–64.
- Mesherov, E. T. (1984): Marker genes in the breeding of muskmelons and water melons. *Nauchnoy tekhnicheskii Bulletin*, VIR, **140**, 13–18.
- Mohr, H. C., Blackhurst, H. T., Jensen, E. R. (1955):  $F_1$  hybrid watermelons from open pollinated seed by use of a genetic marker. *Proc. Amer. Soc. Hort. Sci.* **65**, 389–404.
- Muzik, P., Minh, H. T. (1985): Use of a marker in the heterosis breeding of muskmelons. *Vetömag*, **12**, 4, 29–32.
- Nugent, P. E., Hoffman, J. C. (1974): Inheritance of the halo cotyledon mutant in muskmelon. *J. of Heredity*, **65**, 315–316.
- Robinson, R. W., Munger, H. M., Whitaker, T. W., Bohn, G. W. (1976): Genes of Cucurbitaceae. *Hort. Science*, **11**, 6, 554–568.
- Velich, I. (1985): Use of marker genes in the heterosis breeding of muskmelons. (A marker génnek felhasználása a sárgadinnye heteróznemesítésében.) *Zöldségtermesztési Kutató Intézet Bulletinje*, **18**, 5, 59–64.
- Whitaker, T. W. (1952): Genetic and chlorophyll studies of a yellow-green mutant in muskmelon. *Plant Physiology*, **27**, 203–208.





## THE INFLUENCE OF REPEATED BACK-CROSSES ON THE PRODUCTIVITY OF CYTOPLASMIC MALE STERILE LUCERNE GENOTYPES

B. NAGY

RESEARCH INSTITUTE OF GÖDÖLLŐ, UNIVERSITY OF AGRICULTURE, KOMPOLT, HUNGARY

(Received: 20 November 1986; accepted 20 July 1987)

Lucerne hybrids can be produced using male steriles even under farm conditions. The breeding value of parents with good combining ability selected in the course of progeny tests is of increased significance in crosses between specific genotypes.

The genetic changes caused by repeated back-crosses on the BC<sub>5</sub> lines produced in the first cycle were determined in clone and progeny experiments.

The differences in productivity observed between the clones was reduced by 33% in the BC progeny, indicating that the tendency toward homogeneity is still increasing. In the case of 6 clones and progenies the production of the parent genotypes was proportional to that of their progeny, but one line (BC<sub>6</sub> 109-12/1-4/13-41) gives a clear demonstration of the necessity for progeny tests if gene connections are to be correctly estimated. This opinion was confirmed by progeny — parent covariance analysis.

**Keywords:** Hybrides, inbreeding homogeneity, malesterility, progenytest

### Introduction

The most complicated task in lucerne breeding is to increase the green yield and also the closely correlated hay yield. The differences in productivity observed in breeding experiments are reduced to less than a significant level when synthetic varieties are introduced into general production.

If it is possible to cross specific genotypes, the breeding significance of parents with good combining ability, selected in the cause of progeny testing increases considerably. Lucerne hybrids can be produced on farms, when male steriles are used (Pedersen and Hill, 1972, Böjtös 1973, Nagy, Szalai and Binyneyi 1985). Though the use of large numbers of parents in the progeny tests consumes much time and labour, in special cases such as this, the acquisition of genetic information on the genotype is both necessary and expedient.

The aim of all breeding processes is to avoid inbreeding within varieties intended for general production. Nevertheless, in the breeding process itself, regardless of the basic method (synthetic or hybrid), inbreeding is applied partly to concentrate valuable properties and partly to achieve genetic homogeneity, which is expected to lead to strong direction dominance and epistasy after the crosses have been carried out. Heterosis can only be expected if there is intense depression during inbreeding.



The aim of the current hybrid research was to produce cytoplasmic male sterile inbred lines from which individuals with better combining ability than the original genotype could be selected or from which superior combinations could be bred, by crossing inbred lines.

From the BC<sub>5</sub> lines bred in the first phase, a number of genotypes were selected in order to examine whether the genetic variance had been increased, whether the genotypes, had reached the inbreeding minimum, whether inbreeding depression could still be increased by means of back-crossing, and whether there were any genotypes which were more tolerant to inbreeding during the lengthy process involved in the formation of homozygous lines, or of lines approaching the homozygous state.

### Materials and methods

The influence of inbreeding was examined in the case of male-sterile genotypes. Seven genotypes were chosen at random from mother lines produced with the use of recurrent parents. Of these, one originated from the cms F<sub>1</sub>, one from the cms BC<sub>1</sub> and five from the cms BC<sub>5</sub> generation. In 1982 the parent genotypes were back-crossed to the fertile parent. The female parents needed for the experiment were cloned in winter 1982/83 using the test-tube method evolved in the German Democratic Republic by Steuckardt. Field experiments were carried out in Kompolt on alluvial soil near the River Tarna during spring 1983. In an experiment set up with one factor and 4 replications, the maternal genotypes were each represented by 5 clones per plot and each of their progeny by 24 plants. In the first year the clones were cut 5 times and the progeny 4 times.

In the second year the clones were cut 4 times and the progeny 3 times. In the third year both the clones and the progeny were cut 4 times. The genetic differences between the parent genotypes and the progeny were examined from the point of view of the most important yield components (plant height and shoot number). The significance of the genetic differences was determined using the method described by Sváb (1981) for experiments with one factor and replications. The relative influence of the yield components was determined separately for the parents and the progeny by means of regression analysis using 2 independent variables

### Results

#### *Green yield*

The clones of the female genotypes produced yields ranging from 1423.39 g to 2826.1 g in the 5 cuts made in the year of plantation (1983). The 2 extreme values were reached by BC<sub>5</sub>-109-12(1-4)45-23 and BC<sub>5</sub>-109-8(3-4)57-13, i.e. by individuals stemming from 2 lines with an equal degree of inbreeding. The selective productivity of the genotype which had good productivity in the first year increased further in the second year. In the third year it reached almost the second year. In the third year it reached almost the same level as it did in the second year. The clones of genotypes of the 12(1-4) subline were destroyed in the first replication by the third cutting in the first year, and they proved to have very low productivity in the other replications as well. A more than triple difference, significant at the  $P = 0.1\%$  level, was recorded



between the male sterile genotypes in the course of the 3-year analysis of yield potential. For the property examined, significant differences were shown between the male steriles every year (Table 1).

The plant material used in the second part of the experiment was obtained by repeated back-crosses on the genotypes from the clone experiment. During the 3 years, the difference in relative productivity between the inbred progeny of the highest-yielding male sterile plant BC<sub>5</sub>-109-8(3-4)57-13, gave the biggest green yield in the first year. It exceeded the experimental average by 11-24% each year and took second place among the best inbred progeny with a 121% life productivity, which demonstrates its good persistence.

In the case of the F<sub>1</sub> genotype, back-crossing resulted in about 10% depression in the green yield, but the annual yield dynamics of the BC<sub>1</sub> line

Table 1  
*Relative green yield of male-sterile genotypes of lucerne*  
(Kompolt, 1983-85)

Genotypes	1983	1984	1985	1983-85
F <sub>1</sub> -109	121.64	114.00	127.28	122.61
BC <sub>1</sub> -140-4/2	76.46	71.84	103.00	83.97
BC <sub>5</sub> -109-8(3-4)47-18	91.89	91.33	90.74	91.49
BC <sub>5</sub> -109-8(3-4)57-13	142.45	159.83	158.15	148.78
BC <sub>5</sub> -109-12(1-4)13-24	93.26	105.85	86.25	92.34
BC <sub>5</sub> -109-12(1-4)13-41	71.75	62.14	48.05	63.75
BC <sub>5</sub> -109-12(1-4)45-23	102.54	95.00	86.53	97.04
LSD <sub>5%</sub>	33.19	44.60	61.16	39.81
Mean fodder yield (100%)	1983.86	306.57	979.14	3269.58

Table 2  
*Relative green yield of BC-progenies*  
(Kompolt, 1983-85)

Lines	1983	1984	1985	1983-85
BC <sub>1</sub> -109	101.07	119.21	130.53	114.35
BC <sub>1</sub> -140-4/2 × 09	87.47	109.45	128.56	105.52
BC <sub>6</sub> -109-8(3-4)47-18	104.21	87.58	97.85	99.82
BC <sub>6</sub> -109-8(3-4)57-13	124.10	111.03	119.46	120.79
BC <sub>6</sub> -109-12(1-4)13-24	102.84	89.39	64.22	86.67
BC <sub>6</sub> -109-12(1-4)13-41	120.46	128.81	131.92	125.78
BC <sub>6</sub> -109-12(1-4)45-23	59.85	54.53	27.68	47.08
LSD <sub>5%</sub>	25.71	33.19	65.85	35.47
Mean fodder yield (100%)	669.11	158.86	501.43	1329.40



**Table 3**  
*Analysis of parent-progeny covariance for green yield*  
 (Kompolt, 1983-85)

	FG	SQ <sub>x</sub>	SP	SQ <sub>x</sub>	b	SQ <sub>R</sub>	SQ <sub>E</sub>	FG <sub>E</sub>	F	r
Total	27	42 100 781.86	62 064.51	5 523 427.49						
R	3	8 325 646.58	1 034 515.53	325 528.39						
Genotypes	6	19 678 014.38	1 001 783.48	3 047 987.61	$b_k = 0.0509$	50 999.56	2 996 988.05 (E <sub>3</sub> )			
Error	18	14 097 130.90	94 796.56	1 849 911.49	$b_b = 0.0067$	637.46	1 849 274.03 (E <sub>2</sub> )	17	NS	NS
									0.46	0.1025
Genotypes + Error	24	33 775 145.28	1 096 580.04	4 897 899.10	0.0325	35 602.74	4 862 296.36 (E <sub>4</sub> )	23		

shows good persistence. The greatest depression was found for the BC<sub>5</sub>-109-12(1-4)45-23 genotype: the relative productivity of the BC<sub>5</sub> clone parent was 88.6% over the 3 years, while that of the BC<sub>6</sub> line decreased to 47%. The relative productivity of the genotype BC<sub>5</sub>-109-8(3-4)57-13 which showed outstanding productivity in the clone experiment, decreased by 38% in the progeny. Thus the inbreeding depression was quite substantial despite the good productivity of the BC<sub>6</sub> line.

Improvements in relative productivity were found in many cases. The back-cross progeny of genotype BC<sub>5</sub>-109-12(1-4)13-41 which had the weakest productivity in the clone experiment, had outstanding productivity, and their annual relative yield also increased (Table 2).

*Yield components* (plant height, shoot number) (Tables 3 and 4).

- (1) *Plant height* showed the same tendency as green yield in the case of the parents: F<sub>1</sub> and BC<sub>5</sub> genotypes having good productivity were also tall, and there were no differences between them in the size of the yield component. The significant correlation ( $r = 0.93$ ) indicated the strong connection between plant height and productivity. The order of plant height for the progeny was quite different from that of the parent. Subline BC<sub>6</sub> 109-12(1-4)45-23 which took first place for productivity, came fourth as regards plant height, while the BC<sub>1</sub> progeny of the F<sub>1</sub> genotype grew 25% taller than the average height recorded in the experiment. The mean of the 5 BC<sub>6</sub> lines was 34% less than the average plant height of BC<sub>1</sub> lines produced from the F<sub>1</sub>.
- (2) The same order was found for shoot number as for productivity in the case of the male-sterile parents. A positive divergence was only seen for genotype BC<sub>5</sub>-109-8(3-4)47-18 (green yield 91%, shoot number 111% as a percentage of the experimental mean). Among the progeny, BC<sub>1</sub>-109-106 and BC<sub>1</sub>-140-4/2 × 09 gave the best tillering. These lines which produced

Table 4

*Significance test on the relationship between parents and BC progenies. Green yield*

	SQ <sub>E</sub>	FC <sub>E</sub>	MQ	F
Divergence from linear regression				
Error	1 849 274.03	17	108 722.00 (V <sub>2</sub> )	
Between genotypes	2 996 988.05	5	599 397.61 (V <sub>3</sub> )	5.51 (V <sub>3</sub> /V <sub>2</sub> )
Error + genotypes	4 862 296.36	23		
Hypothesis:				
Corrected $\bar{y}$ -s are the same	3 013 022.33	6	502 170.39 (V <sub>42</sub> )	4.61 (V <sub>42</sub> /V <sub>23</sub> )
b <sub>k</sub> = b <sub>b</sub>	16 034.28	1	16 034.28 (V <sub>432</sub> )	0.15 (V <sub>432</sub> /V <sub>2</sub> )



25% and 20% more than the average, achieved these outstanding yields with a total of 3.4% and 9.1% more shoots.

It can be stated from estimations of covariance, that there is no linear relation between the productivities of the parents and their progeny. (The MQ ratio between genotype  $SQ_R$  and genotypes is insignificant; the F value is 0.85.) The intensity of the progeny-parent regression changed depending on the parent genotypes, so the relation cannot be characterized by a common trend (the F value between the genotypes was very significant).

During inbreeding the clones and their lines fixed different sequences of the genes responsible for productivity. The  $F_1$ , its  $BC_1$  progeny and lines  $BC_5-109-8(3-4)57-13$  and  $BC_6$  could be characterized by greater additivity, while genotype  $BC_6-109-12(1-4)13-41$  had more favourable allele or gene interactions.

Examinations were also carried out to determine whether there were any reliable differences between progeny means ( $\bar{y}$ ) corrected for the common parent grand average by regression within a treatment.

The ratio of variables  $V_{42}$  and  $V_2$  proves, at the  $P = 1\%$  level of significance that yield differences are caused by genetic differences between the progeny.

Plant height was the major yield component in 3-year productivity both for the male-sterile genotypes and for their back-cross progeny. Changes in this property had twice the effect on the yield in the clones, and thrice the effect in the progeny, than changes of a similar order of magnitude in the shoot number. The 2 main yield components showed a very strong significant correlation with green yield. Environmental effects were eliminated as much as possible by taking treatment means as the basis.

### Conclusions

The general laws of inbreeding are valid for populations and lines with a great number of individuals and can only be applied within certain probability limits to genotypes chosen at random. The genotypes in the experiments were chosen without any previous knowledge of their productivity. Data were only available on their male sterility. The 5 individuals from the  $BC_5$  generation represented 2 sublines. The subline based on the plant designated No. 8 from single cross 109 of the  $cms F_1$  was capable of greater, more favourable genetic divergence, judging by the plant progeny, than subline No. 12, 2 plants of which produced less than the experimental mean while a third plant gave only half the yield.

For the planned improvement in hybrid production it is a very favourable sign that an inbred male sterile genotype had outstanding productivity.



This suggest that a wide basis for selection at the genotype level and favourable gene additivity with respect to productivity can be created by means of inbreeding.

In the experiment a relatively narrow genotype composition was examined. The differences between the genotypes can be significantly increased by 5–6 back-crosses with recurrent parents. Thus there is hope that selection on the basis of progeny tests on the genotypes may lead to results.

The genetic homogeneity of the lines (provided their sterility is satisfactory) and the differences between the lines can in themselves make line-selection possible.

Under the joint influence of inbreeding and selection part populations (individuals) having better productivity than the initial material (cms  $F_1$ ) can be produced in inbred lucerne populations. If cytoplasmic male steriles are used for variety production, both additive and non-additive gene effects (dominance, epistasy) can be directly utilized. The line BC<sub>6</sub>-109-12(1-4)13-41 definitely demonstrates the necessity for progeny tests. It was found in general that the productivity of the parent genotype was proportional to that of the progeny, but this line would have been eliminated as a minus variant on the basis of maternal genotype selection, in which case a parent proved by its progeny to be valuable would have been lost.

Selection could have been carried out on the basis of the first-year clone production if only the self-production were important. The progeny-parent covariance analysis and the yield performance of progeny originating from the genotype BC<sub>5</sub>-109-12(1-4)13-41 proved the necessity of progeny test.

In the next phase of this work observations will be made on the crossing effect (heterosis) which can be achieved in the progeny of genotypes showing various degrees of inbreeding depression.

### References

- Böjtös Z. (1973): Lucernatermesztésünk fajtakérdései és a nemesítés helyzete. (Variety problems in Hungarian lucerne — production and the position of breeding.) *Magyar Mezőgazdaság*, **28**, **22**, 9–10.
- Nagy, B., Szalai, Gy., Binnyei, A. (1985): *A KM-Hybridalfa. 23. Időszerű termelési tanácsadó* (KM-Hybridalfa, 23rd Contemporary Production Guide). GATE Kutató Intézetének Kiadványa, Kompolt
- Pedersen, M. W., Hill, R. R. Jr. (1972): Combining ability in alfalfa hybrids made with cytoplasmic male sterility. *Crop Science*, **12**, 500–502.
- Sváb, J. (1981): *Biometriai módszerek a kutatásban* (Biometrical methods in research). Mezőgazdasági Kiadó, Budapest, 407–424.





## INHERITANCE OF SEED COLOUR IN MUSTARD

D. S. RAWAT

DIVISION OF GENETICS, INDIAN AGRICULTURAL RESEARCH INSTITUTE, NEW DELHI, INDIA

(Received: 15 September 1986; accepted 26 August 1987)

A study on the inheritance of seed colour was conducted in different generations derived from crosses between two brown and two yellow seeded cultivars of mustard. It was inferred that the maternal genotype plays the dominant role in the determination of seed colour and this character is controlled by two pairs of duplicate genes ( $R_1$  and  $R_2$ ). The significance of these findings in relation to breeding has been discussed.

**Keywords:** *Brassica juncea*, mustard, seed colour, inheritance

### Introduction

In India, the seeds of commercially cultivated varieties of mustard (*Brassica juncea* L. Czern and Coss) are brown to blackish-brown in colour, but yellow seeds are also available (Sun 1945, Nayar 1968, Vera et al. 1979). Some of the recent studies in the genus *Brassica* revealed that the yellow seeds produce thinner seed coats, lower crude fibres, larger embryos, higher oil content and better meal than the brown (Jonsson and Bengtsson 1970, Stringam et al. 1974, Jönsson 1975, 1977). Keeping in view these merits of yellow seed colour, improvement work for yellow seeded types with high seed yield and oil content was initiated at this institute about a decade ago.

Ample information on the genetics of seed colour in mustard is not available. A knowledge of genetical behaviour of this character would help in formulating more efficient and meaningful breeding programmes. The present study was undertaken with this objective of determining the genetic behaviour of seed colour in mustard.

### Materials and methods

The materials utilized in the present study originated from 2 improved brown seeded cultivars, namely Pusa Bold and Varuna, and 2 yellow seeded cultivars, Dwarf yellow rai and T.M.-8. Each of the 2 brown seeded cultivars was reciprocally crossed with both the yellow seeded cultivars. Four  $F_1$  plants, from each cross combination (excluding reciprocals) were grown in the crossing block. At flowering each  $F_1$  plant was emasculated, back-crossed (BC-1) and test-crossed (TC-1) as female to brown and yellow seeded parents, respectively. Self-pollinated  $F_2$  seeds were also obtained from each  $F_1$  plant by bagging the inflorescences. Sufficient seeds were obtained from every cross-combination, and from all self-pollinated plants.



The seeds of parents,  $F_1$ s,  $F_2$ s, back-crosses (BC-1) and testcrosses (TC-1) were sown in the rabi-season of 1984-85 at the Farm of I.A.R.I., New Delhi, India. The observations of seed colour among their progeny were recorded at maturity. Data were analysed using the Chi-square test.

## Results and discussion

It was observed that crosses with the brown or yellow seeded homozygous female parents bore brown or yellow seeds respectively, regardless of the seed colour of the male parents. This suggested that the seed colour was maternally determined. Earlier studies in mustard did not provide any information on the role of maternal genotypes in seed colour determination (Sun 1945, Nayar and George 1970, Singh and Srivastava 1974, Vera et al. 1979). However, Anand et al. (1985) reported the maternal control of this character which agrees with the conclusion of this study. Contrary to present findings, Heyn (1973) reported that in turnip rape (*Brassica campestris* L.) seed colour is not strictly controlled maternally. This is, however, further supported by the observations of Jönsson (1977) and Schwetka (1982) who noted variations (yellow to black) for seed colour within the same turnip rape plant. The differences between these earlier studies and the one presented here possibly arise from inter-specific variations within the genetic material.

In all the different cross combinations between brown and yellow seeded types,  $F_1$  plants produced brown seeds, which indicated the dominance of brown over yellow. In the present investigation, none of the plants in the  $F_2$ , BC-1 and TC-1 generations bore seeds with different colours on the same plant, as was reported by Jönsson (1977) and Schwetka (1981, 1982) in rape (*Brassica napus* L.) and turnip rape. In the progeny of the  $F_2$  generation, the consistent pattern of segregation of 15 brown: 1 yellow (Table 1), among all cross combinations, clearly revealed that seed colour was governed by 2 dominant genes with duplicate gene action.

In order to confirm the results from the  $F_2$  generation, the segregation pattern was studied in back-crosses and test-crosses. In all 4 crosses, back-cross progenies were brown in colour; and testcross progenies segregated individually into a ratio of 3 brown: 1 yellow (Table 1), as expected. Thus, these results further confirmed the digenic control of seed colour with duplicate gene action. Following the nomenclature proposed by Sun (1945), the 2 dominant genes for brown seed colour have been assigned the symbols  $R_1$  and  $R_2$ . The presence of both the dominant genes  $R_1$  and  $R_2$ , or any one of them either singly or doubly, produced brown seeds. The yellow seeds appeared only when the genotype was recessive at both the loci,  $r_1r_1$  or  $r_2r_2$ .

The results of this study conform with the conclusions of Sun (1945), Vera et al. (1979) and Anand et al. (1985) who reported that, in mustard, seed colours were controlled by duplicate gene action. Though the results of



Table 1

*Genetic behaviour of seed colour in F<sub>2</sub>, BC-1 and TC-1 generations of mustard*

Cross	Generation	No. of progenies*	Seed colour		Ratio	X <sup>2</sup> value	P value
			brown	yellow			
(Varuna × D.Y. Rai)	F <sub>2</sub>	4	1068	65	15:1	0.51	0.30–0.50
(Varuna × D.Y. Rai × Varuna)	BC-1	4	397	—	—	—	—
(Varuna × D.Y. Rai × D.Y. Rai)	TC-1	4	611	194	3:1	0.35	0.50–0.70
(Varuna × T.M.-8)	F <sub>2</sub>	4	838	48	15:1	1.05	0.30–0.50
(Varuna × T.M.-8 × Varuna)	BC-1	4	407	—	—	—	—
(Varuna × T.M.-8 × T.M.-8)	TC-1	4	338	102	3:1	0.77	0.30–0.50
(P. Bold × D.A. Rai)	F <sub>2</sub>	4	906	54	15:1	0.64	0.30–0.50
(P. Bold × D.Y. Rai × P. Bold)	BC-1	4	421	—	—	—	—
(P. Bold × D.Y. Rai × D.Y. Rai)	TC-1	4	375	118	31	0.30	0.50–0.70
(P. Bold × T.M.-8)	F <sub>2</sub>	4	685	39	15:1	0.92	0.30–0.50
(P. Bold × T.M.-8 × P. Bold)	BC-1	4	379	—	—	—	—
(P. Bold × T.M.-8 × T.M.-8)	TC-1	4	369	112	3:1	0.75	0.30–0.50

P. Bold = Pusa Bold

D.Y. Rai = Dwarfyellow rai

\* Because of similar behaviour of individual progenies, the data have been pooled for 4 progenies each.

Sun (1945) agree with the present finding (15 B: 1 Y), he referred to supplementary instead of duplicate gene action, but this does not seem to follow the currently accepted terminology. The description of seeds as purple (Sun 1945), instead of brown, apparently differs from the current description of the pruplish-brown seeds. Vera et al. (1979) also regarded these earlier results to be affected by differences in the colour description and the nomenclature of gene action. However, some results of Vera et al. (1979) did not agree well with the conclusion of this present investigation because they also reported monogenic control, besides duplicate gene action. This partial difference between the two studies may arise from variations in genetic material; the studies of Vera et al. (1979) are based on a European brown seeded cultivar and a Canadian land race and cultivars of brown and and oriental mustard (the brown and yellow-seeded forms of *Bljuncea* L. Coss). Contrary to the present finding, Nayar and George (1979) further Singh and Srivastava (1974) reported monogenic control for this character in mustard. These authors used yellow seeded mutants, in which the probability of achieving two simultaneously mutated loci, is very low due to the irradiation of the brown seeded cultivar Rai-5 Nayar (1968). It seems feasible to assume that the brown seeded line used by Nayar (1968) already had a locus in homozygous recessive condition, and it may explain why Nayar and George (1970) reported the 3 : 1 ratio for seed colour. This assumption, explains why the brown seeded cultivar



RT-11 used by Singh and Srivastava (1974) had a locus in homozygous recessive condition, which resulted in a monogenic inheritance for seed colour.

Furthermore, in turnip rape, for the control of this character, one gene has been reported by Ahmed and Zuberi (1971), two genes by Stringam (1980) and three or more genes by Mohammed et al. (1942), Jönsson (1975) and Schwetka (1982). The latter author also reported the presence of multiple alleles at certain gene loci. Thus, the genetics of seed colour appears to be much more complicated in turnip rape than in mustard. Still, the presence of slightly varying, in distinguishable shades in both brown and yellow seeded forms of mustard requires further investigation.

The knowledge gained in the present study may be profitable used by breeders in more effectively planning the population sizes of breeding materials while developing yellow seeded varieties of mustard, for which in recent years the demand has been growing.

### References

- Ahmed, S. U., Zuberi, M. I. (1971): Inheritance of seed coat colour in *Brassica campestris* L. variety Toria. *Crop. Sci.*, **11**, 309-310.
- Anand, I. J., Reddy, W. R., Rawat, d. S. (1985): Inheritance of seed coat colour in mustard. *Indian J. Genet.* **45**, 34-37.
- Heyn, F. W. (1973): *Beiträge zum auftreten unreduzierter Gameten und zur Genetik einiger Merkmale bei den Brassicaceae*. Ph. D. Thesis, Landw. Fak., University of Göttingen.
- Jönsson, R. (1975): Yellow-seeded rape and turnip. II. Breeding for improved quality of oil and meal in yellow-seeded materials. *Sver. Utsadesf. Tidskr.* **85**, 271-278.
- Jönsson, R. (1977): Breeding for improved oil and meal quality in rape (*Brassica napus* L.) and turnip rape (*Brassica campestris* L.) *Hereditas*, **87**, 205-218.
- Jönsson, R., Bengtsson, L. (1970): Yellow-seeded rape and turnip rape. I. Influence of breeding for yellow seeds upon yield and quality properties. *Sver. Utsadesf. Tidskr.*, **80**, 149-155.
- Mohammad, A., Sikka, S. M., Aziz, M. A. (1942): Inheritance of seed colour in some oleiferous *Brassicaceae*. *Indian J. Genet.*, **2**, 112-127.
- Nayar, G. G. (1968): Seed colour mutation in *Brassica juncea* Hook. f. and Thomas induced by radioactive phosphorous —  $^{32}\text{P}$ . *Sci. and Cult.*, **34**, 421-422.
- Nayar, G. G., George, K. P. (1970): Inheritance of pod arrangement and seed colour in *Brassica juncea* L. Czern and Coss. *Indian J. Genet.*, **30**, 579-580.
- Schwetka, A. (1981): *Samenfarbe bei Kohl und Rüben und deren Einfluss auf die Samenfarbe synthetischer Rapsformen*. Ph. D. Thesis, Landw. Fak., University of Göttingen.
- Schwetka, A. (1982): Inheritance of seed colour in turnip rape (*Brassica campestris* L.). *Theor. Appl. Genet.*, **62**, 161-169.
- Singh, R. N., Srivastava, A. N. (1974): Note on the breeding behaviour of a yellow seeded rai (*Brassica juncea* L.). *Sci. and Cult.*, **40**, 407.
- Stringam, G. R. (1980): Inheritance of seed colour in turnip rape. *Can. J. Plant Sci.*, **60**, 331-335.
- Stringam, G. R., McGregor, D. I., Pawlowski, S. H. (1974): *Chemical and morphological characteristics associated with seed colour in rapeseed*. Proc. 4th Int. Rapeseed Conf., Giessen, FRG, pp. 99-108.
- Sun, P. C. (1945): Genetic studies on *Brassica juncea* Coss. I. Flower colour, leaf shape, seed colour and branching habit. *J. Agri. Ass. China* (Suppl. No.), **50**, 12-13.
- Vera, C. L., Woods, D. L., Downey, R. K. (1979): Inheritance of seed coat colour in *Brassica juncea*. *Can. J. Plant Sci.*, **59**, 635-637.



## STUDIES ON THE AGRONOMIC AND BREEDING POTENTIALS OF SOME INTERSPECIFIC HYBRIDS IN *ARACHIS*

K. O. MARFO\*

AGRICULTURAL EXPERIMENT STATION NYANKPALA, GHANA

(Received: 23 September, 1986; accepted 26 August 1987)

In this study, the breeding and agronomic potentials of thirty-one groundnut lines of interspecific origin (at or near the hexaploid or tetraploid levels) were compared with five standard cultivars. Most of these interspecific hybrids had been selected for their disease resistance. The aim was to compare the cultivars with derivatives of the wild species and to determine to what extent the inclusion of germ-plasm from wild species had increased the variability within the materials. The results indicated that most of the lines derived from wild species performed well in most of the characters studied, the range of variation being often greater than in *Arachis hypogaea* L. Most of these lines are being used in the breeding programs of the International Crops Research Institute for the Semi-Arid Tropics (ICRISAT), especially where disease resistance is one of the main objectives. Nevertheless, there were undesirable characters such as single-seeded pods which resulted in low levels of kernel weight among derivatives of the wild species.

**Keywords:** *Cercosporidium arachidicola* (Hori), groundnut (*Arachis hypogaea* L.), interspecific hybridization, *Phaeoisariopsis personata* (Berk et Curt), *Puccinia arachidis* (Speq.)

### Introduction

The improvement of several traits such as insect and disease resistance, especially the latter in groundnut, otherwise known as peanut; *Arachis hypogaea* L. is difficult. Thus, most of the cultivated genotypes show some amount of susceptibility to these diseases and pests (Stalker et al. 1979). Thus, exploitation of genetic resources from the germ-plasm of wild, closely related species becomes imperative. However, initially there were many problems to be overcome in the hybridization between wild and cultivated species. Two of these problems were the short period available for floral emasculation and the low multiplication rate, the latter probably due to sterile hybrid progenies.

Recent investigations on genome relationships in the section *Arachis* by Singh and Moss in 1982 and 1984 and earlier successful ones by Raman (1973), Gopinath Nair and Raman (1975), Jayaramaiah et al. (1979) and Gibbons et al. (1980) in India, have led to successful solutions for these problems

\* Present Address: Department of Botany and Plant Sciences, University of California, Riverside, California, U.S.A.



Table 1  
Some agronomic traits of the lines evaluated

Treatment and parental genotype	Original species cross	Ploidy level	No. of days to flower (DF)	No. of flowers per plant (NF)	No. of pegs per plant (NP)	Diseases*	
						Cercospora rating	Rust rating
1. H1/1-23-3	<i>A. hypogaea</i> × <i>A. cardenasii</i>	Tetraploid	43	195	55	3.0	4.0
2. H2/5-7-4	<i>A. hypogaea</i> × <i>A. cardenasii</i>	Tetraploid	39	333	155	2.7	4.9
3. H5/CF 19-8	<i>A. hypogaea</i> × <i>A. cardenasii</i>	Tetraploid	39	276	108	3.7	1.5
4. H6/CF 23-4	<i>A. hypogaea</i> × <i>A. batizocoi</i>	Tetraploid	35	348	101	3.3	1.3
5. HP3/15-5-11	<i>A. hypogaea</i> × <i>A. batizocoi</i>	Tetraploid	39	272	113	3.4	1.1
6. 27HP 14-18/9	<i>A. hypogaea</i> × <i>A. batizocoi</i>	Tetraploid	40	211	69	3.8	5.0
7. 29HP 43-1A/5	<i>A. hypogaea</i> × <i>A. cardenasii</i>	Tetraploid	44	142	56	2.7	3.9
8. HP4/16-263-11	<i>A. hypogaea</i> × <i>A. batizocoi</i>	Tetraploid	37	363	137	4.2	1.5
9. H7/17-17-5	<i>A. hypogaea</i> × <i>A. batizocoi</i>	Tetraploid	42	139	54	3.0	3.9
10. 28HP 41/1	<i>A. hypogaea</i> × <i>A. duranensis</i>	Tetraploid	38	118	45	3.4	3.7
11. HP6/3-11-6	<i>A. hypogaea</i> × <i>A. cardenasii</i>	Tetraploid	44	114	45	3.6	4.2
12. HP9/13-101-3	<i>A. hypogaea</i> × <i>A. batizocoi</i>	Tetraploid	34	264	106	4.9	1.1
13. 30-44 × 10017/1	<i>A. hypogaea</i> × <i>A. cardenasii</i>	Tetraploid	44	184	82	3.2	4.8
14. 501-181/67-2	<i>A. hypogaea</i> × <i>A. cardenasii</i>	Tetraploid	37	173	78	4.8	5.1
15. HP3/15-5-11	<i>A. hypogaea</i> × <i>A. batizocoi</i>	Tetraploid	39	235	86	3.0	0.9
16. 27 HP14-18/9	<i>A. hypogaea</i> × <i>A. batizocoi</i>	Tetraploid	43	306	117	5.4	6.3
17. HP2/8-37NPN	<i>A. hypogaea</i> × <i>A. batizocoi</i>	Tetraploid	41	278	49	4.3	1.0
18. HP14/M13-101-6	<i>A. hypogaea</i> × <i>A. batizocoi</i>	Tetraploid	40	285	117	3.8	1.9
19. HP6/7-31-5	<i>A. hypogaea</i> × <i>A. cardenasii</i>	Tetraploid	40	111	34	3.9	5.1
20. H1/2-43-11	<i>A. hypogaea</i> × <i>A. cardenasii</i>	Tetraploid	44	217	57	3.0	4.0
21. H5/CF-19-8	<i>A. hypogaea</i> × <i>A. cardenasii</i>	Tetraploid	37	290	108	3.2	1.7
22. M13 Mutant	<i>A. hypogaea</i> × <i>A. cardenasii</i>	Tetraploid	43	241	87	3.9	4.5

23. <i>A. monticola</i> × M13	<i>A. monticola</i> × <i>A. hypogaea</i>	Tetraploid	40	275	52	3.7	3.9
24. HIC/192/215	<i>A. hypogaea</i> × <i>A. chacoense</i>	Hexaploid	47	175	23	3.1	1.6
25. HIL/8/13/24	<i>A. hypogaea</i> × <i>A. stenosperma</i>	Hexaploid	46	239	42	3.1	3.2
26. HJK8/10/22	<i>A. hypogaea</i> × <i>A. cardenasii</i>	Hexaploid	48	156	33	3.2	1.8
27. H6/5-165-6	<i>A. hypogaea</i> × <i>A. batizocoi</i>	Tetraploid	39	336	177	3.6	3.4
28. H3/4-41-11	<i>A. hypogaea</i> × <i>A. cardenasii</i>	Tetraploid	40	297	136	2.8	1.2
29. H6/5-57	<i>A. hypogaea</i> × <i>A. cardenasii</i>	Tetraploid	45	136	43	3.3	4.0
30. HP43-1A262/CF41-10	<i>A. hypogaea</i> × <i>A. cardenasii</i>	Tetraploid	46	210	58	3.3	3.8
31. H1/4-11-11	<i>A. hypogaea</i> × <i>A. cardenasii</i>	Tetraploid	51	127	33	4.4	4.3
32. TMV2 (Cultivar)	<i>A. hypogaea</i> × <i>Spanish bunch</i>	Tetraploid	34	142	60	8.4	8.5
33. Robut 33-1 (Cultivar)	<i>A. hypogaea</i>	Tetraploid	42	273	156	7.2	8.8
34. M13 (Cultivar)	<i>A. hypogaea</i>	Tetraploid	45	276	111	7.3	7.3
35. Gangapuri (Cultivar)	<i>A. hypogaea</i>	Tetraploid	33	114	65	8.0	7.9
36. Makulu Red (Cultivar)	<i>A. hypogaea</i>	Tetraploid	38	111	77	5.9	7.5
General mean			41	223	81	4.1	3.8
C. V. %			6	25	33	20.0	28
C. D. at 5%			4	92	44	1.4	1.8

\*1 = No disease, 9 = Severe disease.



of interspecific hybridization. In other parts of the world, successes such as those of Conagin and Tella (1972), Sharief et al. (1978), and Smartt and Gregory (1967) have also been very encouraging.

In this investigation, some of those interspecific *Arachis* hybrids at or near the hexaploid or tetraploid level, and which had been selected for a range of characters such as productivity, disease resistance, earliness or combinations of these characteristics were tested along with some established cultivars. The disease resistances were mainly for peanut leaf rust caused by *Puccinia arachidis* Speq. and the early and late leaf spots caused by *Cercospora arachidicola* Hori and *Phaeoisariopsis personata* (Berk et Curt — until recently known as *Cercosporidium personatum* Berk et Curt) respectively.

### Materials and methods

Thirty-six entries including hexaploids and tetraploids of interspecific origin and standard local cultivars were planted during the major season of 1980 at the ICRISAT Research Centre, Patancheru in the Alfisols. The entries (Table 1) were shown in a 6×6 triple lattice design. Each treatment consisted of 3 rows, 4 m in length, spaced 75 cm apart. The plant spacing within a row was 15 cm. Infector rows of alternate plants of 2 cultivars, both highly susceptible to rust and leaf spots were provided between each treatment. The plants were irrigated when necessary to maintain adequate moisture in the soil.

The following pre-harvest records among others were noted:

- (a) Number of days to first flowering.
- (b) Number of flowers produced per day. One plant was selected at random from within each row of a replicate (9 plants per treatment) and the total number of flowers produced daily by each plant was recorded.
- (c) Disease resistance. Scoring for leaf spots and rust on a 9-point scale developed by ICRISAT groundnut Pathologists (Anonymous, 1979).

The post-harvest records on the tagged plants were:

- (a) Percentage of single-lobed pods.
- (b) Pod weight per plant.
- (c) Average kernel weight per plant.
- (d) Average number of kernel per plant.
- (e) Shelling percentage.
- (f) Percentage oil content using Nuclear Magnetic Resonance spectrometer.

### Results and discussion

#### *Flowering*

Significant differences were found to exist between the genotypes as to the number of days they took to flower and their productivity of flowering (Table 1). Significant correlations were also found between the number of flowers produced on one side, and number of pegs, pod weight, number of kernels, and weight of kernels as well as the shelling out-turn (Table 3). The top 9 genotypes with the highest productivity of flowers were all of interspecific origin, mainly derivatives of *Arachis batizocoi* (Krap et Greg. Nom. nud).



Table 2

*Kernel yield and some yield components exhibited by the materials tested*

Treatment	Plant habitus	Percentage single-lobed pods (SSP)	Pod weight (gm/plant) (PWT)	Kernel weight (gm/plant) (WK)	No. of kernel per plant (NK)	Shelling percentage (SP)	Percentage oil content (OC)
2	Spanish erect bunch	23.0	29.7	16.0	58	53.8	50.1
18	Spanish erect bunch	9.7	20.3	11.1	36	54.7	50.9
19	Spanish erect bunch	20.7	2.0	1.4	7	70.0	43.1
32	Spanish erect bunch	8.3	7.3	4.2	22	57.5	44.5
2	Virginia erect bunch	21.3	9.5	5.5	23	61.1	43.3
7	Virginia erect bunch	18.9	3.1	1.4	11	45.2	49.3
8	Virginia erect bunch	11.8	18.4	10.8	42	58.7	46.9
9	Virginia erect bunch	43.6	5.0	3.1	17	62.0	51.2
12	Virginia erect bunch	16.9	12.9	7.3	29	56.6	47.0
15	Virginia erect bunch	31.2	5.9	2.8	17	47.5	42.6
26	Virginia erect bunch	79.7	1.5	0.9	3	60.0	43.7
27	Virginia erect bunch	12.9	14.6	7.9	41	54.1	44.5
28	Virginia erect bunch	31.6	20.3	11.9	41	58.6	48.4
30	Virginia erect bunch	39.2	2.5	1.6	11	64.0	49.1
36	Virginia bunch	8.7	6.4	3.1	12	48.4	47.3
35	Valencia bunch	5.8	12.8	7.5	33	58.6	45.6
1	Virginia Semispreading bunch	35.6	4.2	2.1	19	50.0	49.1
3	Virginia Semispreading bunch	22.9	14.0	8.1	45	57.9	48.5
4	Virginia Semispreading bunch	20.6	18.2	9.1	40	50.0	46.3
10	Virginia Semispreading bunch	25.8	3.6	1.8	11	50.0	33.9
11	Virginia Semispreading bunch	32.5	3.8	2.2	11	57.9	51.2
13	Virginia Semispreading bunch	41.5	8.3	4.6	26	55.4	50.1
20	Virginia Semispreading bunch	21.2	4.3	2.3	18	53.5	50.3
21	Virginia Semispreading bunch	23.3	14.4	7.8	32	54.2	46.4
24	Virginia Semispreading bunch	69.1	2.5	1.4	4	56.9	48.3
29	Virginia Semispreading bunch	42.4	4.1	2.0	15	48.4	53.4
31	Virginia Semispreading bunch	31.8	0.7	0.3	5	42.9	44.0
33	Virginia Semispreading bunch	19.7	38.4	19.6	64	51.0	42.0
6	Virginia Semispreading runner	49.1	5.7	2.7	15	47.4	45.3
16	Virginia Semispreading runner	13.1	24.5	11.9	44	48.6	46.3
17	Virginia Semispreading runner	39.4	4.4	1.7	8	38.6	45.4
22	Virginia Semispreading runner	22.0	11.2	5.3	22	47.3	44.3
34	Virginia Semispreading runner	26.3	21.6	11.3	36	52.3	44.4
14	Valencia Semispreading runner	22.6	13.8	7.1	37	51.5	51.0
23	Virginia runner	63.2	3.4	1.6	6	47.1	44.7
24	Valencia runner	69.1	2.5	1.4	4	56.0	48.3
Mean		29.3	10.6	5.6	24	53.5	46.9
C. V. %		30.6	48.0	4.9	41	18.8	2.6
C. D. (5%)		19.3	8.3	4.5	17	15.7	0.2



**Table 3**  
*Correlation coefficients between some of the variables investigated*

DF	NF	NP	CR	RR	SSP	PWT	NK	WK	OC	SP
DF	-0.27	-0.32	-0.28	-0.04	0.38*	-0.25	-0.34*	-0.27	0.08	-0.22
NF		0.70**	-0.09	-0.30	0.04	0.59**	0.60**	0.59**	-0.12	0.34*
NP			0.10	0.03	-0.31	0.77**	0.82**	0.79**	-0.07	0.33
CR				0.67**	-0.34*	0.30	0.22	0.28	-0.31	-0.02
RR					-0.19	0.20	0.15	0.18	-0.08	-0.02
SSP						-0.29	-0.31	-0.29	-0.04	0.15
PWT							0.94**	0.99**	-0.06	0.30
NK								0.95**	0.04	0.39*
WK									-0.05	0.37**
OC										0.07
SP										

\* Significant at 5% level of probability.

\*\* Significant at 1% level of probability.

*Abbreviation*

*Name*

DF	Days to flowering
NF	Number of flowers
NP	Number of pegs
CR	Cercospora rating
RR	Rust rating
SSP	% of single seeded pods
PWT	Pod Weight
NK	Number of kernels
WK	Weight of kernels
OC	Oil content
SP	Shelling Percentage

### Diseases

For both resistance to *Cercospora* and rust fungi, most of the lines of interspecific origin were highly resistant. Five of such lines (T15, T17, T12, T5 and T28) were almost completely resistant to rust. In addition treatments 28 and 15, which were derivatives of *Arachis cardenasii* (Krap et Greg. Nom. nud) and *Arachis batizocoi* respectively, were found to possess simultaneously high resistances to both leaf spots and rust (Table 1). Similar patterns of resistance have been observed by Stalker et al. (1979), Jayaramaiah et al. (1979) and Kolawole in 1976 from resultant interspecific hybrids. Such lines may be useful for further investigations on mechanisms of resistance to these fungi. It also appears that genes for resistance to *Cercospora* and rust are closely linked, so too the association between *Cercospora* and single-seed poddedness (Table 3).

### Yield and Yield Components

Pod weight indicates that some of the derivatives of wild species such as T2, T18 and T28 (erect bunchy) and T16 are promising future materials in terms of the higher pod yields which they exhibit. Similarly, they performed favorably in terms of the number and weight of kernels produced (Table 2). The relatively wide outside row and narrow inside row spacing used for all the treatments was the optimum in regard to the diverse nature of the lines. Thus the average kernel yield of the bunchy types (about 6 gm) does not seem to be significantly different from the average overall kernel yield of 5.7 gm for the 36 lines.

As expected, the pod weight correlated highly with the number of pegs, kernels and kernel weight (Table 3). Unfortunately, a substantial portion of the lines of interspecific origin developed a single-seed pod, which may be characteristic of wildness. Another factor for which the genotypes derived from wild species showed very high performance was their oil content. T29, which had the highest oil content of over 53%, was followed in descending order by treatments 9, 11, 14 and 18, all being derivatives of wild species. It was also observed that the 7 genotypes with the highest shelling out-turn were all of interspecific origins, with treatment 19 showing as high as 70%.

### Conclusion

The crosses of wild species with cultivated groundnuts, in addition to their increasing the diversity among the genotypes of the resultant progenies, also improved the desirability of the hybrids. For example, it was established that the incorporation of such wild genes raised the disease resistance levels



of the cultivated groundnuts. Furthermore, the desirable trait of average shelling percentage increased in the resultant hybrids, as also did their oil contents. However, the single-lobed characters, which tended to persist in some of the derivatives, and therefore to restrict their yields, presented the major difficulty in completing the desirability of these derivatives of wild species.

Fortunately, the incorporation of some of these wild species into the breeding program of ICRISAT has produced stable, high yielding, foliar disease resistance lines (ICRISAT, 1983, 1984, 1985) which are being made available to various national programs for testing and adoption, or for further use in their various groundnut breeding programs.

### Acknowledgement

I wish to express my sincere gratitude to Dr. J. P. Moss, Groundnut Improvement Program, ICRISAT, for his assistance throughout these studies.

### References

- Anonymous (1979): *Preliminary groundnut foliar diseases assessment trial ICRISAT*, Patancheru, A. P. 502324 A. P. India 27 pp.
- Conagin, C. H. T. M., Tella, R. De (1972): Improving common groundnut (*Arachis hypogaea* L.) by colchicine treatment. *Brazantia*, **31**, (1) 187-198.
- Gopinath Nair P., Raman, V. S. (1975): Cytogenetic relationships and barriers to gene exchange in *Arachis. Oleagineux*, **30**, (10) 419-422.
- ICRISAT (1983): *Annual Report 1982*. Patancheru, A. P. 502324, India.
- ICRISAT (1984): *Annual Report 1983*. Patancheru, A. P. 502324, India.
- ICRISAT (1985): *Annual Report 1984*. Patancheru, A. P. 502324, India.
- Jayaramaiah, J., Siddaramaiah, A. L., Prasad, K. S. K. (1979): Incidence of rust on some species/varieties of groundnuts. *Groundnut Research, University of Agricultural Science, Bangalore* **8** (2), 29-30.
- Kolawole, K. B. (1976): A short progress report on transfer of *Cercospora* resistant traits to the cultivated *Arachis hypogaea* L. *Samaru Agricultural Newsletter*, **18** (1) 40-43.
- Raman, V. S. (1973): Genome relationship in *Arachis. Oleagineux*, **28**, (3) 137-140.
- Sharief, Y., Rawlings, J. O., Gregory, W. C. (1978): Estimates of leaf spot resistance in three interspecific hybrids of *Arachis. Euphytica*, **7** (3), 741-751.
- Singh, A. K., Moss, J. P. (1982): Utilization of wild relatives in genetic improvement of *Arachis hypogaea* L., Part 2: Chromosome complements of species in section *Arachis*. *Theoretical and Applied Genetics*, **61**, 305-314.
- Singh, A. K., Moss, J. P. (1984): Utilization of wild relatives in genetic improvement of *Arachis hypogaea* L., Part 5: Genome analysis in section *Arachis* and its implications in gene transfer. *Theoretical and Applied Genetics*, **68**, 1-10.
- Smartt, J., Gregory, W. C. (1967): Interspecific cross compatibility between cultivated peanut, *Arachis hypogaea* L. and other members of the genus *Arachis Oleagineux*, **22**, 455-459.
- Stalker, H. T., Wynne, J. C., Company, M. (1979): Variation in progenies of an *Arachis hypogaea* × diploid wild species hybrid. *Euphytica*, **28**, 675-684.



## Plant protection

### INCIDENCE OF *TARSONEMINA* SPECIES IN SOIL SAMPLES OF CERTAIN TOMATO VARIETIES

S. M. ABO-KORAH and A. A. YOUNES

PLANT PROTECTION DEPT., FACULTY OF AGRICULTURE, MINUFIYA UNIV.,  
SHEBIN EL-KOM, EGYPT

(Received: 23 March 1987; accepted 17 June 1987)

The occurrence of soil *Tarsonemina* species (*Acari: Heterostigmata*) inhabiting 5 tomato varieties in fields was determined in Minufiya Governorate, Egypt during two successive seasons 1984 and 1985.

Twenty-five tarsonemine mite species belonging to 5 families, were encountered. Of these 5 species were recorded for the first time in Egypt. These are: *Siteroptes sevastianovi* Abo-Korah et Zaki, *Pediculaster Zaheri* Sev. et Abo-Korah, *Pediculaster solimani* Abo-Korah, *Mahunkania hallensis* Rack and *Heterodispus aegyptiacus* Sev. et Abo-Korah. Moreover, 10 *Tarsonemina* species were observed for the first time in tomato fields in Minufiya Governorate.

During the course of investigation, maximum population density of *Tarsonemina* occurred under tomato variety Walter (20 spp. with 32197 inds./m<sup>2</sup>, while the lowest population was found under variety V.F.N. 8 (13 spp. with 6330 inds./m<sup>2</sup>).

One species of the collected mites was recorded only under one tomato variety, but not under the others.

**Keywords:** *Tarsonemina* sp., *Siteroptes sevastianovi* Abo-Korah et Zaki, *Pediculaster Zaheri* Sev. et Abo-Korah, *Pediculaster solimani* Abo-Korah, *Mahunkania hallensis* Rack, *Heterodispus aegyptiacus* Sec. et Abo-Korah

#### Introduction

*Tarsonemina* species may be predators (Smiley and Landwahr, 1976); phytophagous (Suski 1970, 1972 and 1973) or fungivorous (Gurney, Hussey 1967; Suski 1967, 1968; Wicht 1970, Kosir 1975, Abo-Korah 1982) living in soil, litter and organic manure. This supercohort of mites was rather neglected for a long time as it received little attention in Egypt, and from zoologists elsewhere. Many factors such as temperature, moisture, plant cover, microflora, soil type and organic matter, etc., determine to a large degree the composition, nature, distribution and abundance of *Tarsonemina* (Block 1966, Loots and Pyke 1967; Abo-Korah 1982; Abo-Korah and Osman 1983; Abo-Korah and Salem 1982). Because information about the status of *Tarsonemina* occurring among tomato varieties are rather scarce, an attempt was made during the period 1984-1985 to discover the varietal susceptibility to these creatures in the Shebin El-Kom district.



### Materials and methods

The present work was conducted in the Experimental farm of the Faculty of Agriculture at Shebin El-Kom during two successive seasons 1984–1985. Five tomato varieties were cultivated and arranged in a randomized block system of three replicates each. These are: *Lycopersicon esculentum* var. Walter; *Lycopersicon esculentum* var. V.F.N.8; *Lycopersicon esculentum* var. Nagcarlan; *Lycopersicon esculentum* var. Saladette and *Lycopersicon esculentum* var. S.T.D. Peto-94. All agricultural practices were according to the normal system.

Soil samples were taken periodically every ten days by a sampling tool vol. 1000 cc. at two depths, 0–10 and 11–20 cm, under each tomato variety. The extraction of mites was carried out by using the modified Tullgren-funnel. *Tarsonemina* species were mounted on slides by using Hoyer's medium, after treatment with KOH 15% for clearing, then identified to species level according the nomenclature of Sevastianov (1978) and Abo-Korah (1985).

### Results and discussion

Twenty-five mite species of supercohort *Tarsonemina* were collected from soil under five tomato varieties. Of these, 2 species belong to each of the families *Siteroptidae* and *Microdispidae*, 9 species to family *Pygmephoridae*, 7 species of the family *Scutacaridae*, and 5 species to family *Tarsonemidae* (Table 1).

It is of interest to clarify that in the present work, 5 species were recorded for the first time in Egyptian tomato fields. These are: *S. sevastianovi*; *P. zaheri*, *P. solimani*; *M. hallensis* and *H. aegyptiacus*. On the other hand, 10 species were observed for the first time at Minufiya Governorate inhabiting tomato fields (Abo-Korah and Osman, 1980, Kandeel 1981 and Abo-Korah, 1984).

The maximum population density of tarsonemine mites occurred under the tomato variety Walter (32 197 ind./m<sup>2</sup>) during the course of investigation followed by Nagcarlan (16 167 ind./m<sup>2</sup>), while the lowest population occurred under variety V.F.N.8 (6330 ind./m<sup>2</sup>). A similar variation in the population densities were observed by Höller (1962) Abo-Korah (1982), Abo-Korah and Salem (1982) who mentioned that changes in mite densities depend on soil, climatic conditions, vegetation, cultivation practices and biological properties of the species.

Results also showed that the tomato variety Walter seemed to be the most favourable, as it supported the highest number of *Tarsonemina* species (20 spp.), followed by Saladette (18 spp.), Nagcarlan (16 spp.), and 13 species for each of varieties V.F.N.8 and STD-Peto 94. Loots and Ryke (1967) and Abo-Korah (1982) recorded that the host type plays a very important role in distribution and abundance of soil *Acari*. Moreover, one species of the collected mites was recorded only under one variety, but not under the others, i.e. *Bakerdania* sp. occurred only under STD-Peto 94, *T. inaequalis* in Walter field and *S. baculitarsus agaricus* under Saladette only. This may indicate that only one tomato variety.

The species *B. centriger*, *P. mesembrinae* and *H. aegyptiacus* were recorded in maximum population densities during the course of investigation with

**Table 1**  
*Tarsonemina species and density in different tomato varieties*

Species	Average No. of individuals/m <sup>2</sup> (0-20 cm) under tomato varieties					Total
	Walter	V.F.N.8	Nagcarlan	Saladette	STD-Peto 94	
Fam.: SITEROPTIDAE						
<i>Siteroptes priscus</i> Krczal	233	0	33	66	67	399
<i>S. sebastianovi</i> Abo-Korah et Zaki	833	33	333	0	100	1 299
Fam.: PYGMEPHORIDAE						
<i>Bakerdania</i> sp.	0	0	0	0	33	33
<i>B. centriger</i> (Coor.)	13 067	900	6034	2760	1 100	23 867
<i>B. tarsalis</i> Hirst	33	0	400	134	0	567
<i>B. gracilis</i> Krczal	167	33	200	134	133	667
<i>B. gossipia</i> Sev. et Abo-Korah	0	167	0	33	0	200
<i>Pediculaster mesembrinae</i> (Canst.)	12 367	3133	2967	5000	7 133	28 800
<i>P. zaheri</i> Sev. et Abo-Korah	33	0	0	233	0	266
<i>P. solimani</i> Abo-Korah	266	167	66	200	0	699
<i>Mahunkania ballensis</i> Rack	0	0	33	67	100	
Fam.: SCUTACARIDAE						
<i>Heterodispus agyptiacus</i> Sev et Abo-Korah	4 300	2800	4967	434	12 334	
<i>H. Pubescens</i> Mah.	267	433	434	100	33	1 267
<i>Imparipes</i> sp.	33	0	0	0	100	0
<i>Pygmodispus latisternus</i> Pazoli	33	0	0	33	0	66
<i>Scutacarus</i> sp.	67	0	0	0	0	67
<i>S. longitarsus</i> (Berl.)	33	0	33	200	0	266
<i>S. baculiarsus agaricus</i> Norton et Ide	0	0	0	33	0	33
Fam.: MICRODISPIDAE						
<i>Microdispus</i> sp.	33	0	0	0	0	33
<i>Brennandania silvestris</i> Jacot.	66	33	33	234	67	433
Fam.: TARSONEMIDAE						
<i>Tarsonemus</i> sp.	700	266	300	100	333	1 699
<i>T. Waitei</i> Banks	533	66	134	100	200	1 033
<i>T. brevipedes</i> Liv., Mitr. et Shar.	0	33	100	0	0	133
<i>T. inaequalis</i> Liv., Mitr. et Shar.	33	0	0	0	0	33
<i>Lupotarsonemus bilobatus</i> (Suski)	100	66	100	100	33	399
No of species	20	13	16	18	13	
Total	32 197	6330	16 167	10 399	9 733	74 828

L.S.D. between varieties at 5% = 766.4

1% = 1004.9



total population levels of 28 800, 23 867 and 12 334 individuals/m<sup>2</sup>, respectively. On the other hand, the species *Bakerdania* sp., *S. baculitarsus agaricus*, *Microdispus* sp. and *T. inaequalis* were recorded in few numbers (33 ind./m<sup>2</sup> for each). Wood (1960) stated that the plant roots might influence their surroundings of soil *Acari* both physically and chemically, and also by providing organic matter from their dead tissue.

Statistical analysis showed that there were highly significant differences between tomato varieties under study (F. value: 11.05).

### References

- Abo-Korah, S. M. (1982): Interaction between the fungus *Rhizoctonia solani* and certain tarsonemine mite species (*Acari: Tarsonemina*). *Minufiya J. Agric. Res.*, **5**, 401-410.
- Abo-Korah, S. M. (1982a): Occurrence of soil mites under certain cotton varieties in Minufiya Governorate, Egypt. *Ibid.* (in press).
- Abo-Korah, S. M. (1984): *Tarsonemina (Acari: Heterostigmata)* fauna in Minufiya Governorate, Egypt. *Bull. Soc. Ent. Egypte* (in press).
- Abo-Korah, S. M. (1985): A list of new tarsonemine mite species (*Acari: Tarsonemina*) in Egypt. *Proc. 6th Arab. Pest. conf., Tanta Univ.*, 16-17 Sept. 1985. **2**, 321-333.
- Abo-Korah, S. M., Osman, A. A. (1980): Population density of the tarsonemine mites under certain truck crops (*Acari: Tarsonemina; Pygmephoroidae; Tarsonemoidea*). *Minufiya, J. Agric. Res.*, **3**, 387-394.
- Abo-Korah, S. M., Osman, A. A. (1983): The tarsonemid mites under certain field crops in Minufiya Governorate, Egypt. *Bull. Soc. ent. Egypt*, **62**, 191-196.
- Abo-Korah, S. M., Salem, S. E. (1982): Influence of soil temperature and moisture on population dynamics of certain tarsonemine site species (*Acari: Tarsonemina*). *Minufiya, J. Agric. Res.* **5**, 411-420.
- Block, W. (1966): Seasonal fluctuations and distribution of mite populations in Moorland soils, with notes on biomass. *J. Anim. Ecol.*, **33**, 487-503.
- Gurney, B. and Hussey, N. W. (1967): *Pygmephorus* species (*Acarina: Pyemotidae*) associated with cultivated mushrooms. *Acarologia*, **9** (2): 353-358.
- Höller, G. (1962): Die Bodenmilben des rheinischen Lösslechs in ihrer Abhängigkeit von Düngung und anderen Standort-faktoren-Monogr. *Angew. Ent.*, **18**, 44-79.
- Kandeel, M. H. K. (1981): *Ecological and biological studies on some tarsonemid mites*. Ph. D. Thesis, Fac. Agric. Cairo, 307 pp.
- Kosir, M. (1975): Ernährung und Entwicklung von *Pygmephorus mesembrinae* and *P. quadratus* (*Pygme phoridae, Tarsonemini, Acari*) und Bemerkungen über drei weitere Arten. *Pedobiologia*, **15** (5), 313-329.
- SLoots, G. C., Ryke, P. A. J. (1967): The ratio oribatid, Trombidiformes with the reference to organic matter content in soil. *Pedobiologia*, **7**, 121-124.
- Smiley, R. L., Landwehr, V. R. (1976): A new species of *Tarsonemus* (*Acarina: Tarsonemidae*), predaceous on tetranychoid mite eggs. *Entom. Soc. Amer.*, **69** (6), 1065-1072.
- Suski, Z. W. (1967): Tarsonemid mites on apple trees in Poland. IX. *Tarsonemus pauperoseatus* (*Acarina, Heterostigmata*). *Bull. Acad. Pol. Sci. Ser. Sci. Biol.* **15** (5), 267-271.
- Suski, Z. W. (1968): Polish mites of the family *Tarsonemidae* (*Acarina, Heterostigmata*), *Tarsonemus idaeus* *Ibid.*, **17** (10), 637-642.
- Suski, Z. W. (1970): Polish mites of the family *Tarsonemidae* (*Acarina, Heterostigmata*). IV. *Steneotarsonemus gibber*. *Ibid.*, **18** (5), 277-282.
- Suski, Z. W. (1972): Tarsonemid mites on apple trees inq Poland. XI. Field observations on the distribution and significance of *Tarsonemidae* (*Acarina, Heterostigmata*) in apple orchards. *Zes. Prob. Pest. Nauk. Rol.*, **129**, 139-157.
- Suski, Z. W. (1973): A revision of *Siteroptes cerealium* (Kirchner) complex (*Acarina, Heterostigmata, Pyemotidae*). *Ann. Zoologici*, **30** (17), 1-26.
- Wicht, M. C. (1970): Three new species of pyemotid mites associated with commercial mushrooms. *Acarologia*, **12** (2), 262-268.
- Wood, F. W. (1960): Biological antagonism due to phylotoxigenic root exudates. *Bot. Rev.*, **26**, 546-569.



## TOMATO TRANSPLANTING AND SOIL *TARSONEMINA* (ACARI)

S. M. ABO-KORAH and A. A. YOUNES

PLANT PROTECTION DEPT., FACULTY OF AGRICULTURE, MINUFIYA UNIV.,  
SHEBIN EL-KOM, EGYPT

(Received: 29 April 1987; accepted 12 November 1987)

Studies of the relationship between tomato transplanting dates and soil tarsonemine mite species were conducted in Minufiya Governorate during two successive seasons 1984 and 1985.

Soil *Tarsonemina* species varied drastically on the three tomato transplanting dates. The second date (Sept. 18, 1984) of transplanting increasingly affected the species number and their population density (21 spp. — 17 172 ind./m<sup>2</sup>), while on both the first (March 28, 1984) and third (Jan. 2, 1985) planting dates (16 spp. for each) there appeared a lower population, averaging 4153 and 5421 ind./m<sup>2</sup>, successively.

Certain *Tarsonemina* families prefer one transplanting date to another for their activity. This conclusion could be easily deduced also from the species distribution.

**Keywords:** *Tarsonemina* sp., tomato, tomato-transplanting

### Introduction

In Egypt, mites have become serious pests on different truck crops, as favourable weather conditions have allowed a remarkable abundance of the mite fauna to survive and propagate throughout the year.

Many factors such as temperature, humidity soil host type and organic matter determine to a large degree the composition, nature, distribution and abundance of soil acari (Loots and Pyke 1967; Abo-Korah and Salem 1982; Abo-Korah and Osman 1983 and Abo-Korah et al., 1984).

Tomato has occupied an important position among other truck crops and little work has been done on soil *Tarsonemina* species that occur with it. Because of the lack of studies concerning the relationship between the tomato transplanting date and the soil tarsonemine mite species, an effort was made herein to investigate this matter during the seasons of 1984 and 1985 in Minufiya Governorate, Egypt.

### Materials and methods

Random soil samples were taken periodically every ten days by an iron sampler Vol. 1000 cc from tomato fields during the study period from April 1984 to March 1985. Three dates of tomato transplanting were studied; 28 March 1984, 18 September 1984 and 2 January 1985. Mites were carefully extracted by using a Tullgren funnel, then mounted and identified.



Table 1

Effect of transplanting dates of tomato on population density of soil *Tarsonemina* (1984-1985)

	Average No. of mites/m <sup>2</sup> at indicated dates					
	March 28	%	September 18	%	January 2	%
<b>Fam. SITEROPTIDAE</b>		2.41		4.08		1.24
<i>Siteroptes priscus</i> Krczal	67		134		0	
<i>S. sebastianovi</i> Abo-Korah et Zaki	33		567		67	
<b>Fam. PYGMEPHORIDAE</b>		27.02		76.61		82.18
<i>Bakerdania</i> sp.	33		0		0	
<i>B. centriger</i> (Coor.)	500		3 756		3 533	
<i>B. tarsalis</i> Hirst	0		133		100	
<i>B. gracilis</i> Krczal	0		100		189	
<i>B. gossipia</i> Sev. et Abo-Korah	0		0		100	
<i>Pediculaster masembrinae</i> (Canst.)	489		8 878		233	
<i>P. zaheri</i> Sev. et Abo-Korah	0		33		233	
<i>P. solimani</i> Abo-Korah	67		189		67	
<i>Mahunkania hallensis</i> Rack	33		67		0	
<b>Fam. SCUTACARIQUAE</b>						
<i>Heterodispus aegyptiacus</i> Sev. et Abo-Korah	1 933		1 944		233	
<i>H. pubescens</i> Mah.	500		245		33	
<i>Imparipes</i> sp.	33		100		0	
<i>Pygmodispus latisternus</i> Pozoli	33		33		0	
<i>Scutacarus</i> sp.	67		0		0	
<i>S. longitarsus</i> (Berl.)	0		55		100	
<i>S. baculitarsus agaricus</i> Norton et Ide	0		33		0	
<b>Fam. MICRODISPIDAE</b>		1.59		1.36		1.01
<i>Microdispus</i> sp.	33		0		0	1.01
<i>Brennandania silvestris</i> Jacot.	33		233		55	
<b>Fam. TARSONEMIDAE</b>		7.20		3.91		8.82
<i>Tarsonemus</i> sp.	266		350		100	
<i>T. waitei</i> Banks	33		122		211	
<i>T. brevipedes</i> Liv., Mitr. et Shar.	0		67		67	
<i>T. inaequalis</i> Liv., Mitr. et Shar.	0		33		0	
<i>Lupotarsonemus bilobatus</i> (Suski)	0		100		100	
No. of species	16		21		16	
Total	4 153		17 172		5 421	

## Results and discussion

The data presented in Table (1) clearly show that there are notable differences in the population density of soil *Tarsonemina* among the various dates of tomato transplanting. *Tarsonemina* species varied remarkably on the three dates of transplanting. The second date (18 September, 1984) of transplanting showed a more significant increase in mite population density than did the other two dates (17 172 individuals per m<sup>2</sup>). Both the first (28 March

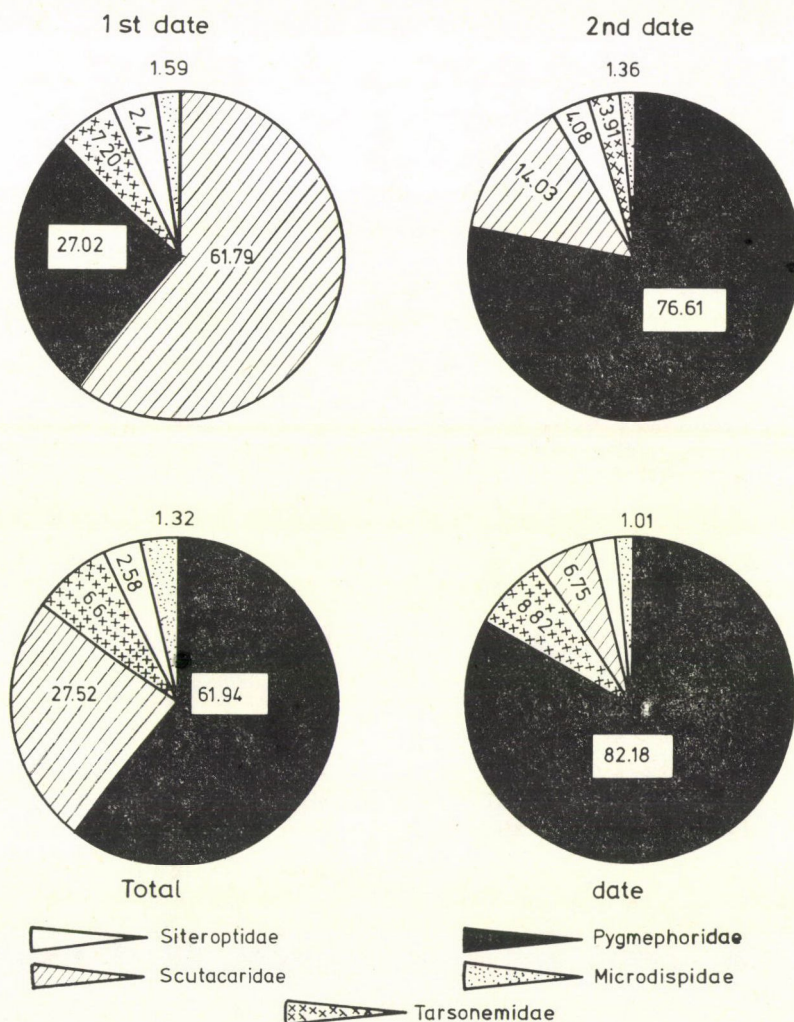


Fig. 1. Relative percentages of *Tarsonemina* families on three transplanting dates of tomato (1984-85)



1984) and third (2 January 1985) planting dates resulted in a lower population, averaging 4153 and 5421 ind./m<sup>2</sup>, respectively.

Soil *Tarsonemina* not only occurred on the second planting date in great number but also dominated qualitatively the other two dates. The total number of species on this occasion were 21 spp., while 16 species were found on each of the other two dates. Loots and Ryke (1967) found that many factors such as temperature, humidity and organic matter determine to a large degree the composition, nature, distribution and abundance of soil animals. Abo-Korah and Salem (1982) stated that the changes as a whole in the population of soil *Tarsonemina* bore strong relationships to the fluctuations of soil temperature and moisture.

The data depicted in Fig. 1 show that on the first transplanting date, *Scutacaridae* were not only found in high density but also their number exceeded those of other *Tarsonemine* families, a total percentage of 61.79, followed by *Pygmephoridae*, *Tarsonemidae*, *Siteroptidae* and *Microdispidae* with percentages of 27.02, 7.20, 2.41 and 1.59, respectively. *Pygmephoridae* flourished on both the second and third transplanting dates constituting 76.61% and 82.18%, respectively. The family *Microdispidae* had the lowest population, amounting to only 1.36% on the first transplanting date and 1.01% on the third date. The number of *Scutacaridae*, *Tarsonemidae* and *Siteroptidae* fell between these two extremes. These results agree with that obtained by Abo-Korah (1984). Such findings lead us to suggest that certain *Tarsonemine* families prefer one definite transplanting date to another in order to carry out their activity. This conclusion can be easily supported also by the species distribution.

The species *H. aegyptiacus* was the most numerous on the first tomato transplanting date (1933 ind./m<sup>2</sup>), while *B. centriger*, *P. mesembrinae* and the former species were represented on the second date by 3756, 8878 and 1944 ind./m<sup>2</sup>, respectively. On the third date *B. centriger* was the dominant species, with a population density of 3533 ind./m<sup>2</sup>. Moreover, certain species of the collected mites were recorded on one date of tomato transplanting but not on the two others. This would indicate that specific species of *Tarsonemina* may reach their peak of activity at certain times of transplanting.

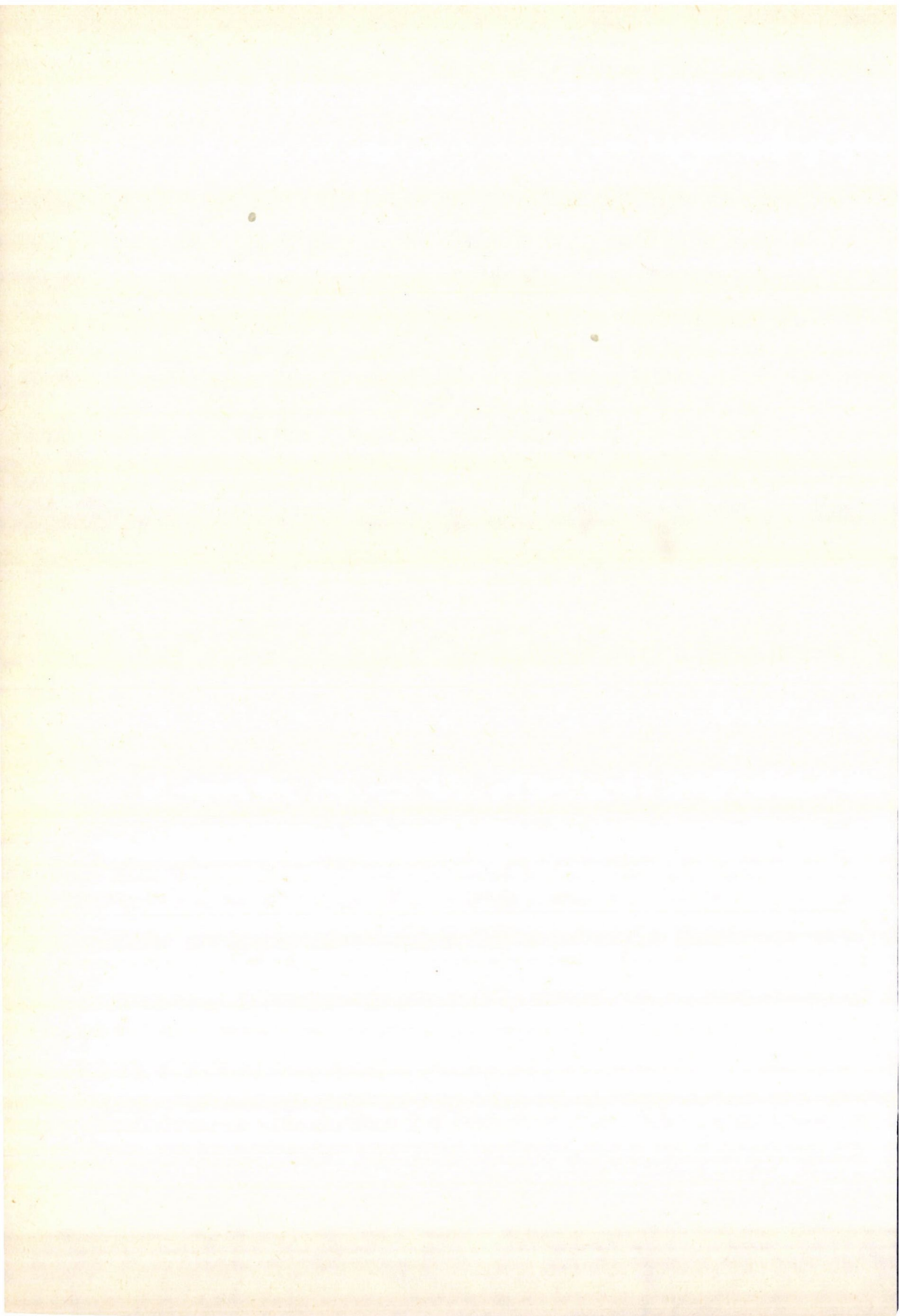
Finally, it can be concluded that there is an interrelationship between the date of transplanting and the presence of various soil *Tarsonemina taraca* as well as their distribution. The date of tomato transplanting affected both the composition and distribution of soil *Tarsonemina*.

### References

- Abo-Korah, S. M. (1984): Survey and population density of the tarsonemine mites under citrus trees in Minufiya Governorate, Egypt. *Bull. Soc. ent. Egypt*, **63**, 13-18.  
Abo-Korah, S. M., Soman, A. A. (1983): Seasonal variations of mite population densities under certain field crops. *Bul. Soc. ent. Egypt* **62**, 197-200.

- Abo-Korah, S. M., Salem, S. E. (1982): Influence of soil temperature and moisture on population dynamics of certain tarsonemine mite species (*Acari: Tarsonemina*). *Minufiya J. Agric. Res.*, **5**, 411-420.
- Abo-Korah, S. M., Radwan, H. S. A., Abo-Elghar, M. R., Salem, S. E. (1984): Occurrence of tarsonemid mites (*Acari: Hetrostigmata*) in different types of soil in Minufiya Governorate, Egypt. *Minufiya J. Agric. Res.*, **8**, 441-447.
- Loots, G. C., Ryke, P. A. J. (1967): The ratio oribatid, Trombidiformes with reference to organic matter content in soil. *Pedobiologia*, **7**, 121-124.





## *Animal production and genetics*

---

### STUDIES ON PROTEIN UTILIZATION FROM ALKALOID-FREE *LUTEUS ALBUS* SEED

JOLÁN JÉCSAI, MARIANNA SZELÉNYI-GALÁNTAI and B. JUHÁSZ

ÁTK. FEEDING RESEARCH INSTITUTE, HERCEGHALOM, HUNGARY

(Received: 14 April, 1987; accepted 15 June 1987)

The parameters determined for alkaloid-extracted sour lupine seed were: biological value 68.1%, total digestibility 79.8%, apparent digestibility 65.7%, net conversion 54.3%, and productive conversion 30.4%. In N-balance tests performed with pigs the daily average N retention was 18.4 g. In pig fattening experiments the groups given alkaloid-extracted lupine meal showed the same rate of feed consumption and body weight increase as the animals feeding on sweet lupine.

**Keywords:** lupine seed meal, alkaloid extraction, protein conversion, fattening results

#### Introduction

Owing to its high protein content the lupine seed is reasonably substituted for imported proteins in animal feed.

In some of our earlier publications (Szelényi-Galántai et al., 1984., Jécsai et al. 1986) the alkaloid and amino acid contents of and protein utilization indices for various lupines grown in Hungary were given.

The utilization of lupine seed meals for feeding purposes is limited by antinutritive substances (alkaloids, glycosides) present in them, as well as by their high fibre- and insufficient methionine- and cystine contents.

To lessen these disadvantageous properties the members of the Department of Agrochemical Technology of the Technical University, Budapest, elaborated a mechanical husking and extraction procedure which favourably influenced the alkaloid- and fibre content of the material of examination.

The experiments were aimed at evaluation the product obtained by chemical and biological analyses, with special regard to the protein utilization of animals.

#### Materials and methods

The alkaloid-free *Luteus albus* meal required for our experiment was obtained from the "II. Rákóczi Ferenc" Co-operative Farm, Vaja.



### Chemical analyses

The nutrient content of the sample was determined as prescribed by the standard MSZ 6830-79. Its amino acid content was measured with a BIO-CAL Typ 200 automatic amino acid analyser.

The alkaloid content was determined according to the standard MSZ 08-1362-80. The digestibility of protein was examined by *in vitro* multienzyme method (Jécsai et al. 1984).

### Biological experiment on rats

The biological value (BV) of the proteins of the examination material, the total digestibility (TD) and apparent digestibility (AD) of protein, further its net (NPU) protein utilization and productive protein utilization (PPU) were established in a N-balance experiment carried out with young male albino rats, as described in works by Bock et al. (1964) and Szelényi-Galántai (1969). The composition of the semisynthetic diet used in the experiment is shown in Table 1.

### Pre-experiment on pigs

The experiment was carried out with 5 barrows of 30 kg average weight. The experiment period was 14 days. The composition of feed is shown in Table 2. After 14 days of feeding 3 animals were slaughtered. The livers were subjected to histopathological examination, and determined for dry matter-, protein-, amino acid- and alkaloid content.

Table 1

Composition of diets prepared with alkaloid-free *Luteus albus* seed (for rats)

Composition	%
Alkaloid-free <i>Luteus albus</i> seed	22.46
Potato starch	52.54
Sunflower oil	10.00
Table sugar	10.00
Vitamin mixture	1.00
Mineral mixture	4.00
Total	100.00

Table 2

Composition of feed mixture in the preliminary experiment with pigs

Designation	%
Maize	20.00
Rye	20.00
Wheat	42.30
Extracted soybean	5.50
Alkaloid-free <i>Luteus albus</i> seed	10.00
L-lysine	0.15
DL-methionine	0.15
Feeding salt	0.40
Feed-lime	0.50
Premix of standard 647.	1.00
Total	100.00

*N-balance examinations of pigs*

The N-balance examinations of pigs were carried out with a method developed at our Institute (Gundel and Babinszky 1978, Gundel et al. 1978, Gundel and Szentmihályi 1979). The composition of feed used in the experiment is shown in Table 3.

*Pig fattening experiment*

The fattening experiment was carried out in the "II. Rákóczi Ferenc" Co-operative Farm, Vaja, with 20 experimental and 20 control animals of mixed sex per group in 2 replications, fed on a prepared composition. The control feed was of similar composition except that it contained yellow-flowered lupine (*Lupinus luteus*) (Table 4).

During the experiments we followed the body weight increase, the specific feed conversion, the health condition and the appetite of the animals.

**Results***Chemical analyses*

*Nutrient content* Table 5 shows the nutrient content of the alkaloid-free *Luteus albus* meal. As seen from the data the crude protein content is remarkably high (42.3%), the crude fibre content is also very favourable (8.3%), and the same can be said of the crude fat content with its 9.4% value.

The total alkaloid content of the alkaloid-free *Luteus albus* with its 0.12% value on dry matter base does not exceed the total alkaloid content of sweet lupines.

**Table 3**

*Composition and calculated nutrient content of feed mixture in N-balance test performed with pigs*

Designation	kg	%
Maize		20.00
Rye		20.00
Wheat		43.80
Extr. soybean		9.00
Alkaloid-free <i>Luteus albus</i> seed		5.00
L-lysine		0.15
DL-methionine		0.15
Feed salt		0.40
Feed lime		0.50
Premix standard 647.		1.00
Total		100.00
DE, MJ/kg	13.78	
Crude protein content		15.30
Starch equivalent	719	
Lysine content total		0.80
Methionine total		0.38
Cystine content		0.24



Table 4

*Composition and nutrient content of feed mixture in the pig-fattening experiment*

Designation	Experimental	Control
	group	
Maize, %	30.00	30.00
Rye, %	10.00	10.00
Wheat, %	43.10	43.10
Alkaloid-free <i>Luteus albus</i> seed, %	5.00	—
Borluta sweet yellow-flowered lupine seed, %	—	5.00
Extr. soybean, %	9.00	9.00
L-lysine, %	0.15	0.15
DL-methionine, %	0.15	0.15
Feed salt, %	0.40	0.40
Feed lime, %	0.50	0.50
Premix, standard 647., %	1.00	1.00
AP-17, %	0.70	0.70
Total	100.00	100.00
DE, MJ/kg	13.75	13.75
Crude protein content, %	15.31	15.50
Starch equivalent, g/kg	721	729
Lysine total, %	0.78	0.78
Methionine total, %	0.23	0.23
Cystine content, %	0.23	0.23

Table 5

*Nutrient- and total alkaloid content of alkaloid-free *Luteus albus* seed*

Analysis	%
Dry matter	89.39
Crude protein	42.32
Crude fibre	8.30
Crude fat	9.42
N-free extr. material	27.29
Ash	2.06
Total alkaloid content, as a percentage of dry matter	0.12 ± 0.06

The chemical analysis of protein digestibility carried out with in vitro multienzyme method gave a 77% value.

*Amino acid composition.* In Table 6. the amino acids contained in the *Luteus albus* sample sent for analysis are listed, in percentage proportion to protein and dry matter, respectively.

Table 6

*Percentage amino acid content of alkaloid-free Luteus albus seed*

Amino acids	% of protein	% of dry matter
Asparatic acid	10.73	4.54
Threonine	4.40	1.86
Serine	5.06	2.14
Glutamic acid	17.80	7.53
Proline	3.92	1.66
Glycine	5.02	2.12
Alanine	4.80	2.03
Cystine	1.19	0.52
Valine	4.76	2.01
Methionine	0.93	0.39
Isoleucine	4.25	1.78
Leucine	8.50	3.59
Tyrosine	4.10	1.73
Phenylalanine	4.41	1.87
Lysine	5.22	2.21
Histidine	2.61	1.13
Arginine	10.01	4.24

The amino acid quantities generally were more favourable under the influence of the treatment than those in the control lupine of the same variety from the previous year (Szelényi et al. 1984).

In the protein of the alkaloid-free *Luteus albus* meal the quantity of sulphur-containing amino acids (methionine + cystine) was 2.12%, and the quantity of lysine 5.22%. On a dry matter base they gave 0.91% and 2.21% values, in the above order of succession.

#### *Biological experiment on rats*

In the case of feeding alkaloid-free *Luteus albus* the average daily N-retention in rats was 39 mg.

The major protein conversion incides calculated on the basis of the N metabolism are given in Table 7. In the alkaloid-free *Luteus albus* meal the biological value of protein was 68.1%, its total digestibility 79.8%, its apparent digestibility 65.7%, and the net- and productive utilization of protein were 54.3% and 30.4%, respectively. These value generally are close to those obtained in our earlier experiments with sweet lupine varieties.

#### *Preliminary experiment with pigs*

During the 14-day experiment the appetite and development of the animals were normal. After 14 days the animals were slaughtered. The dry



matter, protein and alkaloid contents of their livers are shown in Table 8. The average dry matter content was 31.1%, the protein content 21.6%. Alkaloids could not be detected in the livers, nor were histological lesions found.

### *Studies on N-metabolism*

The 4 young pigs examined in the metabolism experiment showed a steady increase of body weight. Their average daily weight gain was 647 g, and they consumed the feed with good appetite.

**Table 7**  
*Percentage trend of protein utilization indices*  
(Experiments with rats)

Indices	In alkaloid-free <i>Luteus albus</i> meal
Biological value	68.1 + 2.9
Total digestibility	79.8 + 5.7
Apparent digestibility	65.7 + 5.9
Net protein utilization	54.3 + 3.9
Productive protein utilization	30.4 + 3.9

**Table 8**  
*Results of livers from pigs consuming alkaloid-free Luteus albus meal*

Serial number of animals	Dry matter %	Protein %	Protein content as a % of absolute dry matter	Alkaloid content
1	29.4	21.5	73.3	0.0
2	30.8	21.1	68.5	0.0
3	30.1	22.2	73.7	0.0
$\bar{x}$	30.1	21.6	71.8	0.0

**Table 9**  
*Daily N-balance data for pigs*

Serial number of animals	N-uptake	N excretion in		N- balance	N- utilization, %
		urine	faeces		
		g			
1	41.59	16.02	6.78	18.79	83.70
2	41.59	16.18	6.48	18.93	84.42
3	41.59	16.30	7.98	17.31	80.81
4	41.59	16.45	6.41	18.71	84.59
$\bar{x}$	41.59	16.24 $\pm$ 0.18	6.91 $\pm$ 0.13	18.43 $\pm$ 0.75	83.38 $\pm$ 1.75

**Table 10**  
*Results of fattening experiments (n = 80)*

Designation	Group	
	Control	Experimental
Initial body weight, kg	28.43	28.90
Final body weight, kg	97.66	99.38
Daily average feed uptake, kg	2.20	2.20
Daily body weight increase during the fattening period, g	543	554
Feed mixture used for 1 kg body weight increase, kg	4.06	3.98

The average daily N-balance data for the pigs are summarized in Table 9. Their average N-uptake was 41.59 g of which they retained 18.43 g. The apparent digestibility of protein was 83.38 g.

#### *Fattening experiments*

The average body weight of the piglets at the beginning of the fattening experiment was 28.43 kg in the control and 28.90 kg in the experimental group; and at the end of the experiment 97.36 kg in the control and 99.38 kg in the experimental group.

During the fattening period the average daily weight increase was 543 g for the control animals and 554 g for the experimental animals. The daily average feed uptake by experimental and control animals was equally 2.20 kg.

For 1 kg body weight increase the control group used 4.06 kg, the experimental group 3.98 kg feed (Table 10).

The health condition, appetite and behaviour of the animals were found to be the same in the two groups.

#### **Discussion**

Fluctuations in the world market prices of imported protein feeds and the strategic character of protein make it necessary to increase the utilization of domestic protein sources.

One of the attempts to achieve this is using the seed of *Luteus albus* — a crop grown with satisfactory yield in some regions of Hungary — as a domestic protein source for partially covering the protein requirements of monogastric animals after a proper treatment.

The treatment of the *Luteus albus* seed, which means removal of alkaloids and a minor degree of husking, changed the values of all components in our material.



The crude protein content was increased by some 10%, while the fibre content was reduced by nearly 4%. As a result of the treatment, the crude fat content also showed a slight increase. It is known (Harvey 1970, Wünsche et al. 1977, Andersen and Just, 1979, Allen 1980) that the amino acid set of the lupine seed is very poor in sulphurous amino acids, and also poor, though less so, in lysine. This was the case with the *Luteus albus* meal used in the present experiment too, but as a response to the treatment an average of 0.5% increase in the quantity of amino acids was observed.

It is important, and it must be emphasized, that the treatment reduced the total alkaloid content from 1.5% to 0.12%, which is hardly more than the alkaloid content in the seed of sweet lupine. This corresponds to the alkaloid content of lupines suitable for feeding purposes.

According to Bélteki and Kovács (1982) the alkaloid content of lupines can be reduced through the improvement of selection (breeding) methods. It will be decided by time and cost factors what method — alkaloid removal and other treatments, or selective procedures — to apply to lessen the anti-nutritive components of the lupine, or both will play some role in it.

According to the experiments with rats the protein conversion indices of the treated lupine meal were favourable, as a whole they came close to the values earlier measured by us in sweet lupines (Szelényi-Galántai et al. 1984).

The treatment resulted in an about 20% better biological value, 18% better net utilization and 17% more favourable productive utilization compared to what we found for the untreated *Luteus albus* seed.

According to the results of preliminary experiments with pigs, the alkaloid-free lupine seed had no unfavourable effect on the appetite and development. The histological examination of livers from the experimental animals did not expose either degenerative or other kind of lesions.

In the N-balance tests the animals consumed the feed with good appetite, their urine and faeces excretion was normal. The average rate of N-uptake and — excretion, the N-balance and the digestibility of protein showed nearly the same values as those obtained earlier when using similar proportions of Nyírségi white-flowered sweet lupine (Szelényi-Galántai et al. 1985).

As proved by the results of the fattening experiment the alkaloid-free *Luteus albus* meal can be used in the feed mixture with as good results as with the sweet lupine. However, the fattening results obtained in the experiment were both in the control and in the experimental group somewhat less than the results of other, similarly arranged experiments (Fekete et al. 1978, Szelényi-Galántai et al. 1985).



## Summary

Experiments were carried out to study the chemical and biological conversion of the alkaloid-free *Luteus albus* meal.

The treatment of the *Luteus albus* seed had a favourable influence on the protein-, amino acid, fibre and fat content of the feed, and reduced the alkaloid content from 1.5% to 0.12%.

According to the N-balance test performed with rats the biological value of the lupine protein was 68.1%, the total digestibility of protein 79.8%, the apparent protein digestibility 65.7%, the net utilization was 54.3% and the productive utilization 30.4%.

In the N-balance test performed with pigs the daily average N-retention was 18.4 g, similar to the value obtained with sweet lupines.

In the pig-fattening experiment, the values of feed consumption, body weight increase and feed conversion were largely the same in the control group given sweet lupine as in the experimental group, where the animals consumed alkaloid-free *Luteus albus* meal.

The chemical and biological conversion of the alkaloid-free lupine seed indicate that it can be used for feeding purposes in the same measure as the sweet lupine.

## References

- Bélteky, B., Kovács, I. (1982): *A csillagfürt* (Lupine). Nyírségi Nyomda.
- Bock, H. D., Nehring, K., Schiemann, R., Hoverka, F., Horzaruk, F., Angelova, L. (1964): Untersuchungen über den Stickstoffumsatz im tierischen Organismus. *Arch. f. Tierernähr. Berlin*, **14**, 13–21.
- Fekete, L., Máray, G., Teér, Gy., Barna-Bukovi, E. (1978): A csillagfürt mint fehérjeforrás a hizósertések takarmányozásában (Lupine as a source of protein in feeding porkers). *Állattenyésztés és Takarmányozás*, Budapest, **27**, (3), 143–157.
- Gundel, J., Babinszky, L. (1978): Különböző takarmányfelvétel és takarmányozás módjának hatása a takarmányok táplálóanyagának emészthetőségére, nitrogén-retenciójára, valamint a hizósertések termelésére (Effect of varying feed uptake and feeding method on the digestibility and nitrogen retention of nutritive substances in feeds, and on the production of porkers. *Állattenyésztési Kut. Int. Közleményei*, Gödöllő, 201–210.
- Gundel, J., Hoffmann, L., Szentmihályi, S., Babinszky, L. (1978): Új típusú anyagcsereketrec növekvő sertések részére és egy többcélú szállító-emelő ketrec (New type metabolism cage for growing pigs, and a multipurpose conveying-hoisting cage). *Állattenyésztési Kut. Int. Közleményei*, Gödöllő, **1**, 305–312.
- Gundel, J., Szentmihályi, S. (1979): A sertések táplálóanyag-szükséglete (Nutrient requirements for pigs) *Magyar Mezőgazdaság*, Budapest, **34**, 35–36.
- Harvey, D. (1970): *Tables of the amino acids in foods and feedingstuffs*. Commonwealth Agricultural Bureaux of Anim. Nutr., Brucksburn Aberdeen, AB2 9SB Scotland.
- Jécsai, J., Szelényi-Galántai, M., Juhász, B. (1984): Fehérjék emészthetőségének vizsgálata *in vitro* multienzim módszerrel (Study of protein digestibility by *in vitro* multienzyme method). *Állategészségügyi és Takarmányozási Közlemények*, Budapest, **4**, 241–249.
- Jécsai, J., Szelényi-Galántai, M., Juhász, B. (1986): A különböző csillagfürtfajták antinutritív hatása a patkányok fehérje-anyagcseréjére (Antinutritive effect of various lupine varieties on the protein metabolism of rats). *Állategészségügyi és Takarmányozási Közlemények*, Budapest, *Phylaxia*, **2**, 91–98.
- Szelényi-Galántai, M., Jécsai, J., Juhász, B., Bódis, L. (1984): Sárga- és fehérvirágú édes csillagfürt, valamint szójafajták takarmányozási értékének megállapítása (Determination of the feeding value of yellow and white flowered sweet lupine and of soybean varieties). *Állattenyésztés és Takarmányozás*, Budapest, **33**, (3) 281–288.
- Szelényi-Galántai, M., Jécsai, J., Juhász, B. (1984): Lehetőségek csillagfürt-magfajták fehérje biológiai értékének javítására (Possibilities of improving the biological value of seed protein in lupine varieties). *Állattenyésztés és Takarmányozás*, Budapest, **33**, (4) 371–378.
- Szelényi, E. (1969): Nitrogénforgalmi vizsgálatok a takarmányfehérjék biológiai értékének meghatározására (Nitrogen balance tests to determine the biological value of feed proteins). *Állattenyésztés*, Budapest, **18**, (2) 189–191.
- Szelényi-Galántai, M., Jécsai, Gy., Juhász, B. (1985): Szójafehérjék helyettesítése különböző édes csillagfürt fajokkal sertések takarmányában (Replacement of soybean protein by



various sweet lupine species in the feed of pigs). *Állattenyésztési és Tak. Kut. Int. Közleményei*, Gödöllő, **1**, 535-541.

Wünsche, J., Bock, H. D., Kreinebring, G., Wiese-Müller, W., Becker, J. (1977): *Aminosäure-Gehaltstabelle 5*. Dummerstorf-Rostock.

MSZ 0830-1979: Takarmányok táplálóértékének megállapítása (Determination of the nutritive value of feedstuffs), Budapest.

MSZ 081362-80: Édes csillagfürt ipari takarmánygyártás céljára (Sweet lupine for industrial feed production), Budapest.

## Lectures

### RECENT RESULTS IN MELON BREEDING\*

K. MOZSÁR

UNIVERSITY OF HORTICULTURE AND FOOD INDUSTRY, BUDAPEST, HUNGARY

(Received: 13 May 1987; accepted 20 July 1987)

Both species of melons constitute a very modest proportion of the total vegetable production in Hungary. The sowing area of watermelons and muskmelons fluctuates between 8000 and 10 000 ha annually, of which only 800-1000 ha are occupied by muskmelons (Fig. 1).

Their market value increases if there is a substantial yield loss in orchards due to frost damage. In such cases they play a greater role in the summer fruit supply. The annual marketing value of melons is around 950-1200 million forints.

The technology of melon production has changed little from traditional methods in the greater proportion of the sowing area, though large-scale methods of production have undergone a substantial development recently. It is likely that, simultaneously with the choice of varieties, this too will contribute to increases in both marketing value and profitability.

The other important factor in successful production is the variety cultivated. Plant breeding carried out over the past ten years has provided farmers with standard and  $F_1$  varieties possessing greater genetic potential for both species.

The first Hungarian successes appeared at much the same time as foreign breeding results and originated partly from the rich local population stock, and partly from the use of foreign varieties, hybrids and mutants. After the first watermelon hybrids of the seventies (K heterosis, K triploid), the triumph of the hybrids "Szigetcsépi-51", "Hevesi Fu-to" and "Gömb Fu-to" over the standard varieties counted as a real success (Fig. 2).

For watermelons the current varieties have many properties which should be maintained, such as the dark rind, which must be thin but strong, the bright red, soft flesh, the small seeds, the flavour and aroma.

Progress is needed, however, in regard to yield potential to an increase in the sugar content and to an improvement in the resistance to certain specific diseases, e.g. to *Fusarium*, anthracnose, powdery mildew and viruses.

\* Presented at the "János Lippay" Scientific Session held on November 20th 1986.



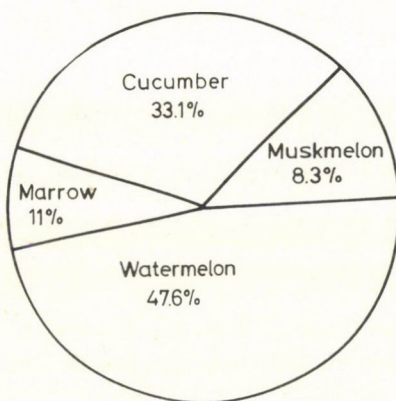


Fig. 1. Ratio of various species of *Cucurbitaceae* on the 17060 ha growing area

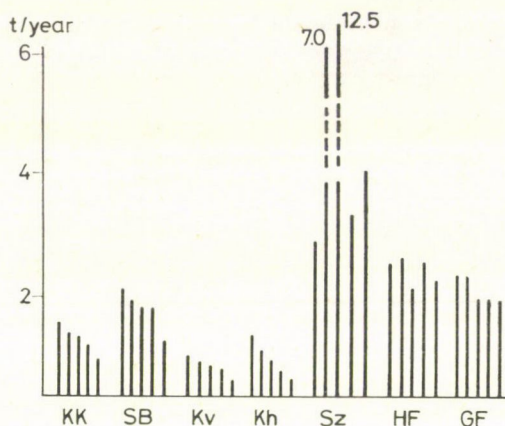


Fig. 2. Watermelon seed sales (1981–1985) Marketed varieties: Korai kincs (Kk); Sugar Baby (SB); Kecskeméti vöröshúsú (Kv); Kecskeméti heterózis (Kh); Szigetsépi-51 F<sub>1</sub> (Sz); Hevesi *Fusarium* tolerant (HF); Gömb *Fusarium* tolerant (GF)

For muskmelons, too, earliness, the intensive green colour of the flesh, its soft consistency, its flavour and aroma are properties which must be retained.

Improvements must be made in yield potential, transportability and sugar content. Characters awaiting introduction are intensive yellow flesh colour with the flavour and aroma that accompany it. Here, too, better disease resistance is needed against anthracnose, *Fusarium*, powdery mildew and viruses.

The extremely varied application of *breeding methods* has already produced results.

— Selection from local varieties resulted in new forms as long ago as the sixties. These include the muskmelon varieties “Ezüstananász”, “Javí-

tott-Zentai", "Turkesztán", etc. and the watermelon varieties "Hevesi", "Szentesi", "K-vöröshúsú", "Korai kincs", etc.

- Due to the long period required for the testing of progeny generations, combination breeding is time-consuming, so it led to no new varieties in the case of watermelons, but resulted in the muskmelon varieties "Tétényi-cseres", "Líra" and "Dixi".

*Heterosis breeding* has proved the best method of melon breeding in Hungary. Researchers at the Department of Plant Genetics and Breeding began their search for parent partners with good combining ability in the sixties. In the course of analyses on a number of partial diallel crosses, several  $F_1$  hybrid combinations for both watermelons and muskmelons proved to be of practical use with respect to earliness, yield potential, quality and disease resistance (Figs 3 and 4; Table 1).

Apart from the success of the hybrid variety "Szigetcsépi-51", mention should be made of the suitability of "Hungária-8" for early production, the

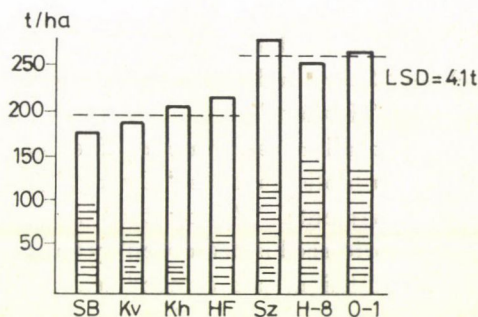


Fig. 3. Yield potential of the varieties (1986). Varieties examined: Sugar Baby (SB); Kecskeméti vöröshúsú (Kv); Kecskeméti heterózis (Kh); Hevesi *Fusarium* tolerans (HF); Szigetcsépi-51  $F_1$  (Sz); Hungaria-8  $F_1$  (H-8); Orosházi-101  $F_1$  (O-1)

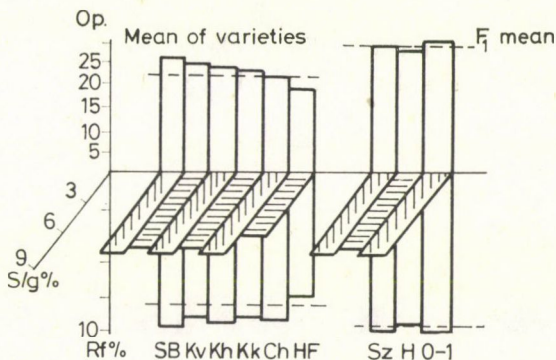


Fig. 4. Quality of the varieties (1986). Varieties examined: Sugar Baby (SB); Kecskeméti vöröshúsú (Kv); Kecskeméti heterózis (Kh); Korai kincs (Kk); Charleston-H (Ch); Hevesi *Fusarium* tolerant (HF); Szigetcsépi-51  $F_1$  (Sz); Hungaria-8  $F_1$  (H-8); Orosházi-101  $F_1$  (O-1)



**Table 1**  
*Fusarium resistance of the varieties (1985)*

Variety	% Withering			Index
	Isolates			
	A	B	C	
Sugar Baby	61	64	73	66
Kecskeméti vöröshúsú	43	47	51	47
Kecskeméti heterózis	41	45	47	44
Korai kincs	43	52	59	51
Charleston	18	22	26	22
Hevesi	28	33	29	32
Szigetcsépi-51	35	38	40	37
Hungária-8	26	34	39	33
Orosházi-101	22	30	38	30

The isolates used in the provocative inoculations originated from Heves (A), Békés (B) and Montfavet France (C). The values presented indicate seedling sensitivity

favourable earliness, yield potential and quality of "Korall", the concentrated ripening and high yield of "Star" and the excellent quality of the late-ripening but high-yielding variety "Kobalt".

In the case of muskmelons a number of early, high-yielding, good quality  $F_1$  hybrids are available, such as "Gyöngy", "Borostyán", "Ázúr" and the late-ripening experimental "Aranygömb".

The production of hybrid seed is promoted by the fact that all the maternal lines are monoic. In addition, non-divided leaf markers are utilized for two hybrids in the case of watermelon, while xantha (yellow cotyledon and foliar leaf) markers have been developed for three maternal muskmelon lines.

This is complemented by the introduction into the seed production technology of the use of a gametocide containing ethephon (CEPA) as active agent, shown by examinations carried out so far to be suitable for reducing the high manual labour costs involved in hybrid seed production and thus for the creation of more favourable prices for  $F_1$  seed.

## INFLUENCE OF ETOLOGICAL FACTORS IN ESTABLISHING THE GENETIC VALUE OF MECHANICAL MILKABILITY\*

L. SZAJKÓ

UNIVERSITY OF AGRICULTURAL SCIENCES, MOSONMAGYARÓVÁR, HUNGARY

(Received: 22 April 1986; accepted 16 June 1986)

It is a well known fact that the milking speed of cows, which is a hereditary trait, influences the time of machine milking, the extent to which the udder is emptied and the occurrence of mastitis cases.

It is therefore important to determine the milking capacity of cows. Environmental factors, however, may influence the cow's behaviour, thus affecting the realistic estimation of milking speed.

According to the results of instrumental examinations, the different methods of preparation and post-milking, as well as circumstances which disturb the cows, may cause a change in their behaviour.

For example, when warm water (36 °C) was used for washing down, quiet behaviour and a tightening of the udder were observed, while when cold tap-water was used the cow stepped sideways and the udder took longer to tighten. The contraction of the udder and the upward movement of the milk in the milk-ducts have been verified by X-ray examinations. In the course of instrumental examinations it was observed that on the appearance of a stranger the cow followed the movement with its eyes. When more than one person appeared wearing usual clothes, the cow followed their movements by turning its head; and when all this was accompanied by loud talk, the cow's movements became more intensive, spreading to the neck and even to the body.

What did the instrument show parallel with the cow's behaviour?

The Uberograf 3, an apparatus that registers the quarters of the udder and the speed of milking, shows the quantity of milk accumulating in the course of milking in the form of a diagram (for each quarter of the udder).

- If only one person was present there was little or no change in milk flow. The diagram did not show more than 6-10 seconds of disturbance.
- If more than one person was present a 5-18 second disturbance in milking was observed.

\* Lecture held at the 4<sup>th</sup> Seminar "Angewandte Nutztier Ethologie an der Bayer Landesanstalt für Tierzucht" 17 October 1985 Grub (BRD).



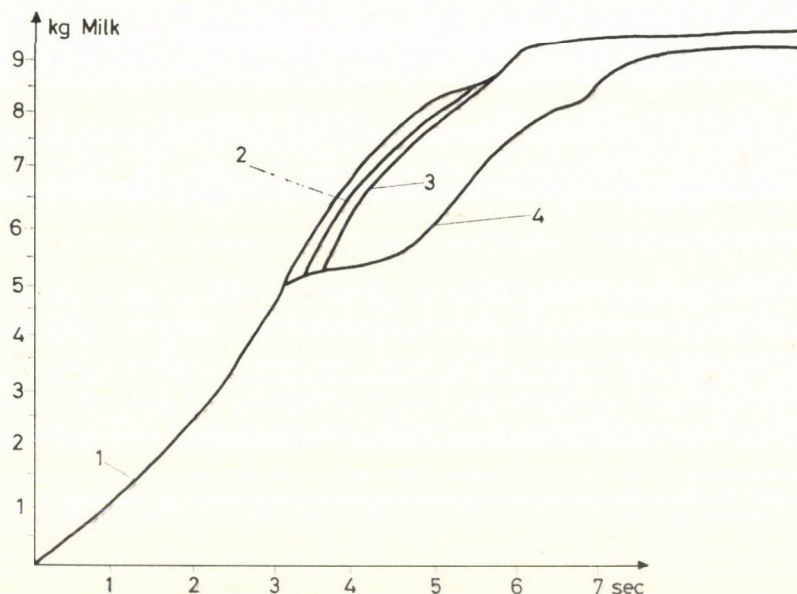


Fig. 1. Effect of disturbing factors of different intensities on milking (Averaged for 5 milkings of the same cow) (1) Control; (2) On the appearance of an unusually dressed person with strange behaviour the cow stops giving milk for 8 seconds. (3) On the appearance of several persons with unusual clothes and behaviour the cow stops giving milk for 15 seconds. (4) When a number of noisy strangers appear milking is suspended for 80 seconds

— Loud conversation conducted by several persons caused a 30–110 second stagnation in the milk flow.

These three cases cannot be regarded as circumstances causing an alarm reflex; nevertheless, the changed behaviour led to disturbances in the milk flow.

In the milk platforms on large-scale farms, for example in cross-type milking platforms, disturbing circumstances evoked the strongest responses from cows near the strangers, while those farther off felt protected, so their behaviour and milk flow did not change.

The change in behaviour and its disturbing effect on milking was more intensive in the case of sensitive cows, which were better milkers.

All this suggests that the genetic value of the milking speed cannot be determined simply by the kg/minute value for the whole milking period.

The instrumental measurements demonstrated that changes in behaviour caused by different methods of preparation and post-milking and by various disturbing circumstances had the least effect when milking took at most 1 minute. Thus, when determining the genetic potential the maximum milking time must be taken into consideration, as this parameter is hardly influenced by the different behaviour patterns.

Great precision is required when several cows are milked simultaneously. The time of post-milking and the manner in which it is carried out are of considerable importance. It often happens that the same milker returns to the same cow at quite different times on various occasions of milking.

From the main phase of milking to post-milking a shorter or longer period of idle milking can be observed.

What is the cow's response to different periods of idle milking? In the case of 20-30 seconds even 40 seconds of idle milking the behaviour of the cow did not change. When idle milking lasted for a minute or so the cow looked expectant and the carriage of its head changed. When idle milking continued, the cow attempted to leave the milking platform.

Increasing reactions, moving away when the teat cups are pulled, backward glances or even kicks call attention to sharp pains in the udder.

As a conclusion it can be established that even the "micro-changes" observed in the behaviour of cows during milking are of great importance, as they have an import on milk yields and influence the determination of the genetic value of milk egestion. Thorough observations render it possible to preserve the healthy condition of the udder.

All in all, the milking conditions must be such that behavioural disturbances will not occur in up-to-date livestock farms.





## *Review*

---

# DEVELOPMENTS IN THE TRACE ELEMENT RESEARCH OUR KNOWLEDGE AT PRESENT AND TRENDS IN THE NEAR FUTURE

I. PAIS

UNIVERSITY OF HORTICULTURE AND FOOD INDUSTRY

(Received: 11 March 1988)

W. Mertz wrote in his paper (1980) that in the recent history of nutrition the first half of our century was characterised as the vitamin era, and the second part of this century will be known as the era of trace elements.

Till 1940 — according to the rigid rules of Arnon and Stout (1939) — the science accepted only 7 elements as essential: iron, zinc, manganese, cobalt, copper, molybdenum and iodine but in the sixties — thanks to the development of analytical accuracy — some “new” essential trace elements came into the family of the physiologically important trace elements: e.g. chromium, nickel, vanadium, selenium, etc. This development gave birth to the problem of searching possibly the total periodical system: to discover which more new elements can have the rank of a physiologically important or promotive element?

In recent some years, many new results came in the focus of scientific awareness that a lot of elements which were earlier regarded as “toxic heavy metals”, their biological importance and sometimes their essentiality nowadays were fully demonstrated. We would like to accentuate that this problem is only the question of concentration level! We can learn from the very old chemist and pharmacist, Paracelsus, who wrote that the toxicity is only the problem of concentration.

If we evaluate the problem that from the 88 stable elements which constitute the geophysical inorganic matter how many elements can participate in the life processes? We can disclose only the noble gases (these elements under common circumstances do not form compounds!) and some strongly radioactive elements. To summarize: about 77 elements may possibly have the ability to become full member of biosphere as macro- or micro-nutrients. This opinion is demonstrated in Figure 1, which contains 11 biogen and macro-element (their symbol is not signed in the figure): H, O, C, Na, K, Mg, Ca, N, P, S and Cl.



The micro-elements — as the figure shows — are classified in 5 groups.

(1) The group of "classically essential" trace elements: manganese, molybdenum, iron, cobalt, copper, zinc and iodine.

(2) The group of partly essential trace elements: vanadium, chromium, nickel, boron, selenium and fluorine.

(3) The group of physiologically promotive trace elements: lithium, titanium, silicon, tin and bromium.

(4) The group of elements of which promotive role is partly verified: rubidium, caesium, strontium, tungsten, platinum, cadmium, aluminium, gallium, germanium, lead and arsenic.

(5) Finally the greatest group of trace elements, the promotive role of which — under special circumstances — is conceivable: beryllium, barium, scandium, yttrium, zirconium, hafnium, niobium, tantalum, rhenium, ruthenium, osmium, rhodium, iridium, palladium, silver, gold, mercury, indium, thallium, antimony, bismuth, tellurium and the rare earths. This excludes the synthetically-produced promethium.

Returning to our basic problem, the development and importance of the trace element research — underlined the aspects of human life — may be classified under three headings.

#### *New results in the physiological role of the trace elements*

The development of physiology and biochemistry demonstrated very clearly that enzymes and enzyme-systems play a key biological role. These systems may be classified in two groups: the first being the group of metallo-enzymes which contain as integrated component, usually one (but in some cases more) trace element. The number of these chemically known metallo-enzymes may be estimated at over one thousand and it is well understandable that using the most modern methods of isolation and also the most sensitive analytical methods, the known number of these type enzymes will grow very rapidly in the next some years.

The second and nowadays much greater group of enzymes is that for whose activation trace elements are needed. In other words it means that the activity of the enzyme is insufficient to fulfill its physiological role without the given trace element.

These two groups of enzymes display a great difference. In the first group the role of the trace element is generally specific: that means, in most cases, it is impossible to be replaced by another trace element. In the second group the replacing is relatively common: usually a new metal-ion with the same charge, and with a similar ion-radius, can replace the mean activating metal-ion.

This knowledge gave us many practical opportunities. In the agricultural



## PHYSIOLOGICAL IMPORTANCE OF TRACE ELEMENTS

I <sub>1</sub>	II <sub>1</sub>	III <sub>2</sub>	IV <sub>2</sub>	V <sub>2</sub>	VI <sub>2</sub>	VII <sub>2</sub>	VIII <sub>2</sub>	I <sub>2</sub>	II <sub>2</sub>	III <sub>1</sub>	IV <sub>1</sub>	V <sub>1</sub>	VI <sub>1</sub>	VII <sub>1</sub>	VIII <sub>1</sub>	
<div>☐ Generally essential</div> <div>▤ Partly essential</div> <div>▨ Physiologically promotive</div> <div>▩ Promotive physiological role partly verified</div> <div>▪ Promotive physiological role conceivable</div>																
Li	Be									B				F		
										Al	Si					
		Sc	Ti	V	Cr	Mn	Fe	Co	Ni	Cu	Zn	Ga	Ge	As	Se	Br
Rb	Sr	Y	Zr	Nb	Mo	Tc	Ru	Rh	Pd	Ag	Cd	In	Sn	Sb	Te	I
Cs	Ba	★	Hf	Ta	W	Re	Os	Ir	Pt	Au	Hg	Tl	Pb	Bi	Po	
		★★														
★ La Ce Pr Nd Pm Sm Eu Gd Tb Dy Ho Er Tm Yb Lu																
★★																

Fig. 1. Physiological importance of trace elements

production the supply with trace elements is one of the most important factors of plant production and of the animal feeding. When the given trace element is either deficient or in surplus stage, in both cases we can calculate with some diseases and as a consequence of them, with decrease of the agricultural production.

*The contamination of human environment*

As a consequence of modern human life, the earlier developed and for a long time practically the same natural equilibrium started to change dramatically. The first source of it is the industrial production. If we calculate first the energy-production, it is combined with emission of a great amount of sulphur-dioxide to the atmosphere and also some industries plants and automobiles emit different nitrogen-oxides in the air. The consequence of these processes is the acidic rain, which produces many of negative results in the environment (corrosion of old monuments, etc.). One of most dangerous consequences of the acidic rain is the change of the soil's pH value. If the common pH value decreases, the change in the uptake of different nutritive elements is also tremendous. For example, the solubility of aluminium, iron and manganese



(and some other elements) may increase on a large-scale. These facts have some very important consequences:

(a) Earlier the scientific opinion had seen the aluminium as a non-toxic element. But now we know that some diseases (possible the Alzheimer-disease etc.) may have a connection with the higher aluminium-level of the human body. It is also conceivable that some animal, or plant disorders are in correlation with the greater Al-concentrations within the biosphere. Without any doubt, that aluminium, if it is in higher level that is common, must have an antagonistic effect upon some essential or promotive nutrient elements.

(b) Parallel with the decrease of soil pH, the manganese concentration (and also the cadmium-levels etc.) will be increased, which has a toxicity consequence for the whole food-chain.

(c) In the recent literature we can find data that the so-called sudden infant death syndrome (SIDS) is also in connection with the higher  $\text{SO}_2$  content of the air and this component has a more dangerous effect, if the selenium-content is lower than is required.

(d) The contamination of nature causes tremendous problems of human health. We would now mention the cadmium, lead, mercury and arsenic concentrations, which can be dangerous for all parts of the food-chain. As we can see in the recent literature the Itai-itai disease in Japan, the Minamata-disease also in Japan, and some other worldwide disorders are consequences of the massive concentrations of these elements.

We have not mentioned some other trace elements (beryllium, thallium, etc.) which were earlier not members of the biosphere but now these elements have an increasing chance to enter the food-chain. Till now we have not enough knowledge about their physiological role and therefore the preventive methods are also not quite clear regarding these new health problems.

### *The problem of civilisation and immunological diseases*

We are convinced that the "civilisation diseases" and in connection with them, new immunological problems of the living organisms — including plants, animals and human beings — are intimately connected with the trace element supply of the food-chain. In the most modern literature we can find some hundred publications which deal with the biochemical-nutritional basis of different cancer-types, heart infarction and some other diseases (Wilson-disease, Alzheimer-disease, multiple sclerosis, etc.).

We have not enough data to say that this or that disease has only a nutritional background, or the primary cause is the deficiency or surplus level of this or that element. The analytical data convincingly show, that in these cases some element may be in a minus or plus relation. From these facts it is not possible to draw the conclusion that these deficiency or toxicity symp-



toms themselves have a causative correlation with the disease, but from day to day an increasing number of convincing results may be the basis of analytical diagnosis or in some cases the basis to give the missing quantity of the element or diminish the surplus of it.

The most convincing and well-known example is the Keshan-disease in China. As reported in the appropriate literature, about 300 million people have suffered with heart-muscle disease and from year to year a growing number of population have become new victims. Lastly — approximately 2 decades ago — it was clearly demonstrated that the cause of this is the very low dietary uptake of selenium. That is, without enough selenium the glutathione-peroxidase enzyme is not able to fulfil its duty, to eliminate the surplus inorganic and organic peroxides, and therefore they can destroy the membranes of the heart-muscles. Similar problems may be found throughout Scandinavia and also in New Zealand.

If we can believe the modern theories of cancer diseases, their biochemical mechanism is based on the activity of cell-oxidation processes carried out by peroxide radicals. The glutathione peroxidase enzyme activated by selenium is able to destroy these radicals. Therefore selenium is nowadays believed to be one of most potential agents against various cancers.

Based on the facts or ideas mentioned above, at least some hundred publications stated that some cancer diseases may be prevented or chemotherapeutically cured by application of different selenium-compounds. The greatest discussion on the application form is: should inorganic or organic selenium-compound be applied? We have enough experiences to say that the organically bound selenium (bound to proteins, or as selenium-containing amino acids, like selenomethionine, selenocysteine, etc.) seems to be more effective, but sometimes the selenite or selenate forms also provide good results.

We have never believed that the selenium is a universal panacea, but we — together with a great number of scientists — are convinced that selenium has much more importance than has been previously accepted.

Going on to the questions of immunology, we would like to recall a phrase mentioned in 1986 at the International Conference in Munich as the main topic for discussion of the next Conference: "Not the cure, but the prevention of diseases!" This pronouncement has a deep meaning: we can and we should like to give more and more energy to the naturally existing immunological systems to prevent the diseases by the own basis! We do not agree with the use of different antibiotics and other drugs, which are — in general — dangerous for the future of humanity in too high levels. They do not provide the ability to fight against the diseases, to develop the own immunological defensive system, and these drugs are rather the sources of weaknesses in the immunological systems.



## THE CONCEPT OF THE INTER- DISCIPLINARY TRACE-ELEMENT RESEARCH

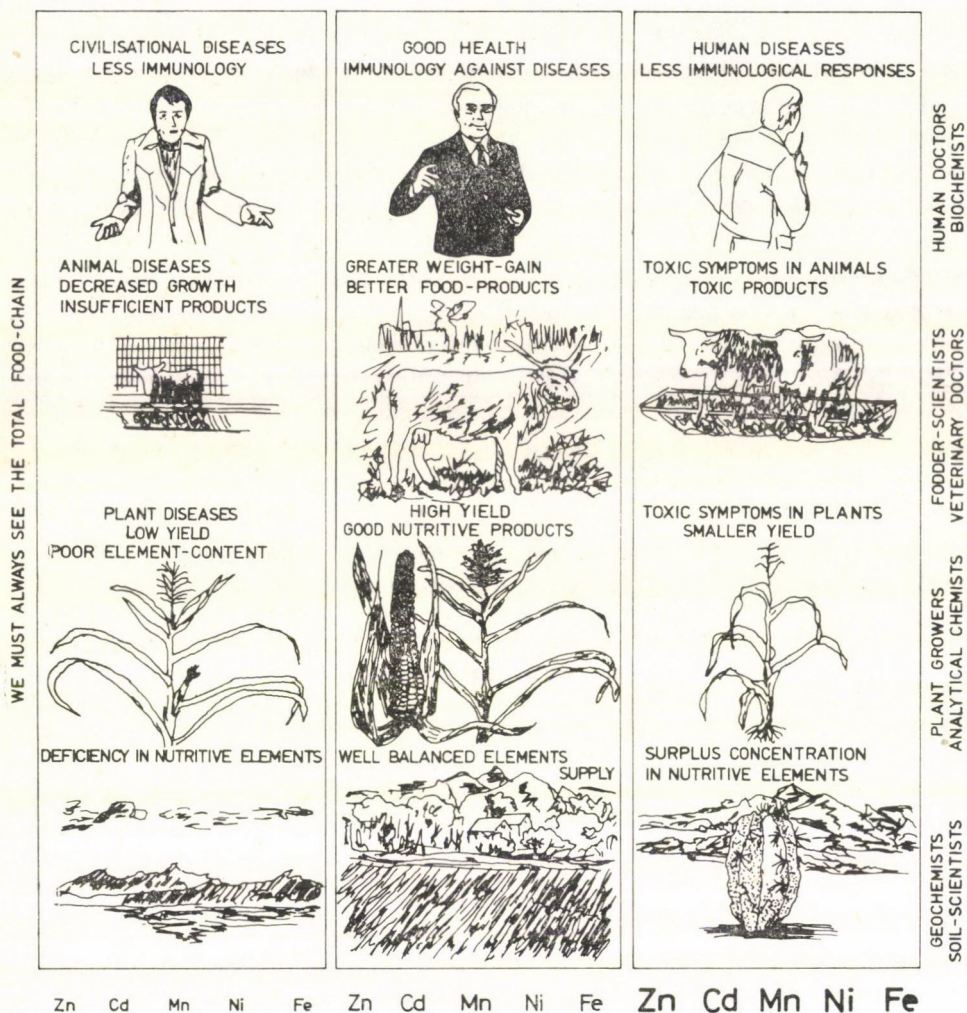


Fig. 2. The concept of the interdisciplinary trace element research

It is also acceptable from the recent trace element literature, that some trace elements — for example zinc — are connected with the aging. Nobody can believe that by application of the most modern biochemical methods and of more healthier nutritional ways, we can wage a successful fight against death, but we believe that applying the appropriate trace element supply



in the elderly, we can slow the aging processes and in acceptable good health. The most important point is to maintain a healthier life style, which is antagonistic to smoking, drinking too much, having a more "comfortable" life without movement, without physical efforts.

### *Solution of the problems of trace element research*

Regarding the problems of trace element research, we can reach only one conclusion: life and the food-chain are intimately connected. Therefore, if we would like to take any good steps to improve the trace element supply in this or that field, we can do it only if we accept the main idea, that the trace element problem is a uniform one, which can be searched if we see the whole spectrum of the food-chain. In other words, we must determine the way of trace elements from their geochemical sources to human health. The importance of the interdisciplinary trace element research can be seen in the Figure 2.

This figure represents that geochemists and soil scientists; the agricultural scientists dealing with the questions of plant and animal nutrition; the veterinary and human doctors; the biochemists and analytical chemists ought to combine their knowledge and their efforts. We may accentuate that we will only in this way be able to solve the most important questions of the trace element research!

This interdisciplinary work may be organised in one country as we have done it in Hungary: the Trace Element Committee of the Hungarian Academy of Sciences which combined the scientists of 6-8 different scientific fields to solve the common problems of trace element research. But only one small country, with such little resources as Hungary, can not solve the main problems of humanity. Therefore we need, and this need always increases to combine our scientific efforts worldwide.

As we demonstrated earlier, the health of plants and animals, in other words the level of agricultural production, is in deep correlation with the plant and animal nutrition, with the activity of different enzymes and enzyme-systems: with the optimal supply of different trace elements. Human health is also directly or indirectly dependent upon the daily trace element uptake of our body. The future of humanity is connected with the prospective results of the trace element research. On this basis, we ought to conclude that besides other scientific fields, the trace element research is one of the most important for the future of the human race.

### References

- Aalbers, Th. G., Houtman, J. P. W. (1985): Relationships between trace elements and atherosclerosis. *Sci. Total Environm.* **43**, 255-283.  
Al-Tawil, N. G. (1985): *Immunological studies in patients with nickel, cobalt and chromium sensitivity*. Doct. Dissertation, Stockholm, 1-53.



- Anderson, R. A. (1986): Trace elements and cardiovascular diseases. *Acta Pharmacol. Toxicol.* **59**, 317-324.
- Arnon, D. L., Stout, P. R. (1939): The essentiality of certain elements in minute quantity for plants with special reference to copper. *Plant Physiol.* **14**, 371-375.
- Balducci, L., Wallace, C., Khansur, T., Vance, R. B., Thigpen, T., Hardy, Ch. (1986): Nutrition, cancer and aging: an annotated review. I. Diet, carcinogenesis and aging. *J. Am. Ger. Soc.* **34**, 127-136.
- Beguín, Y., Weber, G., Delbrouck, J. M., Roelandts, I., Robaye, G., Bury, J., Fillet, G. (1986): Serum trace elements during chemotherapy for acute myelogenous leukemia. *Acta Pharmacol. Toxicol.* **59** (7), 270-273.
- Blazsek, I., Máthé, G. (1984): Zinc and immunity. *Biomed. Pharmacother.* **38**, 187-193.
- Bogden, J. D., Oleske, J. M., Munves, E. M., Lavenhar, M. A., Bruening, K. S., Kemp, F. W., Holding, K. J., Denny, T. N., Louria, D. B. (1987): Zinc and immunocompetence in the elderly: baseline data on zinc nutriture and immunity in unsupplemented subjects. *Am. J. Clin. Nutr.* **46**, 101-109.
- Boysen, F. (1987): *Maternal and postnatal immune deficiency related to magnesium, copper and zinc depletion*. Proc. VI. UOEH Internat. Symp., Kitakyushu (Eds: Brown, S. S. and Kodama, Y.), 39-40.
- Carpentieri, U., Myers, J., Thorpe, L., Daeschner III. C. W., Haggard, M. E. (1986): Copper, zinc, and iron in normal and leukemic lymphocytes from children. *Canc. Res.* **46**, 981-984.
- Chandra, R. K. (1985): Trace element regulation of immunity and infection. *J. Am. Coll. Nutr.* **4**, 5-16.
- Chandra, S., Chandra, R. K. (1986): Nutrition, immune response, and outcome. *Progr. Food Nutr. Sci.*, **10**, 1-65.
- Chiyoia, S., Hanada, K., Hashimoto, I., Katabira, Y. (1987): *Influence of zinc deficiency and cell-mediated immunity*. Proc. VI. UOEH Internat. Symp., Kitakyushu (Eds: Brown, S. S. and Kodama, Y.) 37-38.
- Chowdhury, B. A., Chandra, R. K. (1987): Biological and health implications of toxic heavy metal and essential trace element interactions. *Progr. Food Nutr. Sci.*, **11**, 55-113.
- Davies, B. E., Anderson, R. J.: (1987): The epidemiology of dental caries in relation to environmental trace elements. *Experientia* **43**, 87-92.
- Ehmann, W. D., Markesbery, W. R., Alauddin, M., Hossain, T. I. M., Brubaker, E. H. (1986): Brain trace elements in Alzheimer's disease. *Neuro Toxicol.* **7**, 197-206.
- Fell, G. S. (1986): Trace element metabolism in chronic renal failure: update and perspectives. *Nephrol.* **6** (3), 26-29.
- Garfinkel, D. (1986): Is aging inevitable? The intracellular zinc deficiency hypothesis of aging. *Med. Hypoth.* **19**(2), 117-137.
- Hardy, Ch., Wallace, C., Khansur, T., Vance, R. B., Thigpen, J. T., Balducci, L. (1986): Nutrition, cancer and aging: an annotated review II. Cancer cachexia and aging. *J. Am. Ger. Soc.* **34**, 219-228.
- Heck, D., Ochs, A., Klempnow, A., Maier, K. P., Kratt, C. (1987): Localized changes in trace element concentrations within diseased human liver lobules. *Nucl. Instr. Meth. Phys. Res. B* **22**, 196-200.
- Husami, T., Abumrad, N. N. (1986): Adverse metabolic consequences of nutritional support: micronutrients. *Surg. Clin. N. Amer.* **66**, 1049-1069.
- Huston, R. K., Shearer, T. R., Jelen, B. J., Whall, P. D., Reynolds, J. W. (1987): Relationship of antioxidant enzymes to trace metals in premature infants. *J. Parent. Ent. Nutr.* **11**, 163-168.
- Iesato, K., Ueda, S., Wakashin, M., Wakashin, Y., Yoshida, H., Kato, I., Mori, Y., Mori, T., Ogawa, M., Nishimura, N., Yamamoto, S., Ohta, Y., Okuda, K. (1987): *Is routine urine examination enough for monitoring the nephrotoxicity in environmental exposure of heavy metals? A study of "Minamata-disease" — chronic poisoning of organic mercury*. Proc. VI. UOEH Internat. Symp., Kitakyushu (Eds: Brown, S. S. and Kodama, Y.), 341-342.
- Johansson, E., Lindh, U., Johansson, H., Sundström, C. (1987): Micro-PIXE analysis of macro- and trace elements in blood cells and tumors of patients with breast cancer. *Nucl. Instr. Meth. Phys. Res. B* **22**, 179-183.
- Kromhout, D. (1987): Essential micronutrients in relation to carcinogenesis. *Am. J. Clin. Nutr.* **45**, 1361-1367.
- Kwiatek, W. M., Cholewa, M., Kajfosh, J., Jones, K. W. (1987): Correlation of trace elements in hair of patients with colon cancer. *Nucl. Instr. Meth. Phys. Res. B* **22**, 166-171.
- Leonard, T. K., Mohs, M. E., Ho, E. E., Watson, R. R. (1986): Nutrient intakes: cancer causation and prevention. *Progr. Food Nutr. Sci.* **10**, 237-277.



- Mertz, W. (1980): *Implication of the new trace elements for human health*. Proc. 3. Spurenelem. Symp., Jena (Eds: Anke et al.) 11-15.
- Mooradian, A. D., Morley, J. E. (1987): Micronutrient status in diabetes mellitus. *Am. J. Clin. Nutr.* **45**, 877-895.
- Nan, B. S., Li, C. S., Chen, L. H. (1986): Significance of low levels of blood and hair selenium in dilated cardiomyopathy. *Chin. Med. J.* **99**, 948-949.
- Nestler, J. E., Clore, J. N., Failla, M. L., Blackard, W. G. (1987): Effects of extreme hyperinsulinaemia on serum levels of trace metals, trace metal binding proteins, and electrolytes in normal females. *Acta Endocrin.* **114**, 235-242.
- Niskanen, J., Marniemi, J., Piironen, O., Maatela, J. Mäki, J., Vuori, I., Seppänen, A., Kallio, V., Aromaa, A. (1986): Trace element levels in serum and urine of subjects died in coronary heart disease. *Acta Pharmacol. Toxicol.* **59**, 340-343.
- Nomura, A., Heilbrun, L. K., Morris, J. S., Stemmermann, G. N. (1987): Serum selenium and the risk of cancer, by specific sites: case-control analysis of prospective data. *J. Natl. Canc. Inst.* **79**, 103-108.
- Pais, I. (1985): *Some aspects of the development in the research of the hardly known trace elements and the importance of the interdisciplinary trace-element research work*. Proc. Internat. Tr. El. Symp., Budapest, (Ed.: Pais, I.) 3-14.
- Pais, I., Fehér, M., Nagy, B., Papp, Kl. (1987): *Effects of titanium on other nutritive elements*. Proc. VI. UOEI Internat. Symp., Kitakyushu (Eds: Brown, S. S. and Kodama, Y.), 135-136.
- Pais, I. (1988): *The importance of hardly known trace elements and convincing data on the beneficial character of titanium*. Proc. Internat. Plant Physiol. Congress, New Delhi (under publication)
- Prohaska, J. R. (1987): Functions of trace elements in brain metabolism. *Physiol. Rev.* **67**(3), 858-901.
- Proudfost, F. G., Hulan, H. W., McRae, K. B. (1987): The effects of dietary micronutrient, fat and protein components in palleted feeds on the incidence of sudden death syndrome and other traits among male broiler chickens. *Can. J. Anim. Sci.* **64**, 159-164.
- Ringdal, O., Andersen, K. J., Svendsen, E., Julshamn, K. (1986): Trace elements and myocardial infarction, an autopsy study from western Norway. *Acta Pharmacol. Toxicol.* **59**, 358-360.
- Salonen, J. T., Alfthan, G., Huttunen, J. K., Puska, P. (1984): Association between serum selenium and the risk of cancer. *Am. J. Epidemiol.* **120**, 342-349.
- Salonen, J. T. (1986): Selenium and human cancer. *Ann. Clin. Res.* **18**, 18-21.
- Salonen, J. T. (1987): Selenium in ischaemic heart disease. *Internat. J. Epidemiol.* **16**, 323-328.
- Sandstead, H. H. (1968): A brief history of the influence of trace elements on brain function. *Am. J. Clin. Nutr.* **43**, 293-298.
- Schrauzer, G. N., Molenaar, T., Mead, S., Kuehn, K., Yamamoto, H., Araki, E. (1985): Selenium in the blood of Japanese and American women with and without breast cancer and fibrocystic disease. *Jpn. J. Canc. Res.* **76**, 374-377.
- Schrauzer, G. N. (1987): *Effects of selenium antagonists on cancer susceptibility: new aspects of chronic heavy metal toxicity*. Proc. VI. UOEI Internat. Symp., Kitakyushu, (Eds: Brown, S. S. and Kodama, Y.), 91-98.
- Vernie, L. N. (1984): Selenium in carcinogenesis. *Biochem. Biophys. Acta.* **738**, 203-207.
- Virtamo, J., Valkeila, E., Alfthan, G., Punsar, S., Huttunen, J. K., Karvonen, M. J. (1987): Serum selenium and risk of cancer. *Canc.* **60**(2), 145-148.
- Vruwink, K. G., Keen, C. L., Gerschwin, M. E., Hurley, L. S. (1987): Studies of nutrition and autoimmunity. Failure of zinc deprivation to alter autoantibody production when initiated in disease-established mice. *J. Nutr.* **117**, 177-192.
- Wallwork, J. C. (1987): Zinc and the central nervous system. *Progr. Food Nutr. Sci.* **11**, 203-247.
- Willett, W. C., Stampfer, M. J. (1986): Selenium and human cancer. *Acta Pharmacol. Toxicol. Suppl.* **59**(7) 240-247.
- Yin, S., Pingsheng, L., Runhua, Z., Guilin, L., Zhengxin, Z., Yinkun, F., Guodong, L., Guiquin, S., Shizhen, W. (1987): Determination of trace elements in hair of Wilson's disease patients using PIXE. *Nucl. Instr. Phys. Meth. B* **22**, 191-192.





## Book reviews

---

*Information on maize* (Informatsionnyi byulleten' po kukuruze. — Koordinatsionnyi Tsent. Sev po probleme KOTS-2. Nauchno-issledovatel'skii Inst. S/h. Van Martonvásár. No. 4. 1985).

This 421-pages publication (Edited by the Agricultural Research Institute of the Hungarian Academy of Sciences, Martonvásár, as the coordinator of maize research for the CMEA countries) contains 33 papers reporting the results of wide-ranging research aimed at improving maize and carried out within the framework of the CMEA KOTS-2 programme. The 21 papers in the *first chapter* concentrate upon the plant pathogen and animal pests currently causing the greatest damage to maize. The 6 papers in *Chapter 2* discuss breeding maize for quality, and the major methodological questions related to this topic while a further 6 papers (*Chapter 3*) are concerned with the increase of production and yields. The greater part of the book is devoted to the discussion of plant pathological problems. In some cases, due to the different conditions and current problems facing the various socialist countries, the subjects are presented from contrasting viewpoints. The book can, in fact, be considered as a synthesis of the most topical subjects in maize research and breeding, including the most recent results and current opinions on these problems.

Among the diseases affecting maize, the authors deal mainly with stalk and ear *Fusarium* (*F. graminearum*, *F. culmorum*, *F. moniliforme*, *F. oxysporum*) and other

pathogens causing stalk rot (Drimal, Galejev et al., Borovskaya et al., Klucskó et al., Kizmus 1984), but several authors (Dragonics, Palaversics et al., Milatovics et al., Navratskaya et al., Pencsics et al. 1984) also study other pathogens (e.g. *Ustilago* sp., *Sorosporium* sp., *Helminthosporium* sp., *Nigrospora* sp., *Colletotrichum* sp.). Investigations were carried out on the physiological genetic, breeding and other agronomic aspects of resistance (Ivanovich et al., Rafalski et al., Toldi and Petrics, 1984). Several authors, describe the direct and indirect yield-reducing effects of tissue damage (Ivakhnenko et al., Galejev et al., Klucskó et al. 1984) and the differences in resistance between lines and hybrids (Galejev et al., Klucskó et al., Drimal 1984), while others investigate the inheritance of resistance (*Ustilago* sp.) in SC-TC-DC hybrids and BC combinations (Marton, Szundy and Kovács 1984, Klucskó et al., Navratskaya et al. 1984) or the pleiotropic nature and correlation of the trait (Klucskó et al. 1984). Many authors emphasize the priority of breeding, particularly in the case of early hybrids (Ivakhnenko et al., Langauerovna et al., Drimal 1984). Several authors describe artificial and natural methods for the evaluation of breeding stock and varieties (Navratskaya et al., Galejev et al., Kizmus 1984). These authors emphasize the role of provocative agrophons (agronomical conditions). They do not consider natural infection sufficient for evaluational purposes and thus they stress the need for joint artificial and natural conditions, describing various scales for the evaluation. Navratskaya



et al. (1984) particularly emphasize the role of the provocative conditions elaborating a complex immunity method. Monocultures, dense sowing and a plentiful N supply are regarded as favourable conditions for mass infection. Other plant pathogens and pests causing damage to maize are also dealt with from the standpoint of plant breeding and the methodology of artificial infection (Navratskaya, Inglik and Csizmár 1984). Among the maize pests, resistance to *Ostrinia nubilalis* is discussed by Longauerovna et al. 1984, while the evaluation of resistance to *Rhopalosiphon* sp. and other pathogens, including work on research methodology, is dealt with by other authors (Rabinchuk et al., Milatovich et al. 1984). Galejev et al. (1984) indicate the connection between *Fusarium* infection and quality (BC, Inbred, Opaque-2). Nevertheless, on the basis of a study on VIR lines, the possibility of combining resistance and high lysine content in a single genotype is included. Several authors deal with the agronomic and economic evaluation of damage (Galejev et al., Navratskaya et al., Ivanovich et al. 1984). On studying the correlation between N forms ( $\text{NO}_3$ ,  $\text{NH}_4$ ) and infection, Toldi (1984) concludes that the pathogenicity of the fungus is not influenced by the N quantity alone.

As a result of this research, various lines can be distinguished for their better resistance and good combining ability: F/2, 502, W401, A417, W64/A, Oh-29, Co 152; others as having better resistance only: HMV 404, OK-104, Od-306, rF-7, A-632 (Klucskó et al. 1984); and others as being more resistant to leaf spottedness: BC-10, MO-17, H-99, W37A, W64/A (Milatovich et al. 1984). More resistant hybrids have also been produced, such as the OdMV-310 hybrid developed at Martonvásár and Odessa in cooperation (Klucskó et al. 1984), the joint Czechoslovak-Hungarian hybrid TaMv 310, state registered in 1984 (J. Longauerovna), and the registered hybrid 423 L-t (Rotár et al. 1984). Navratskaya et al. (1984) also report the production of new hybrids capable of yielding 0.8–1.8 t/ha more than the standard (Dnepropetrovski 430).

The second chapter is concerned with breeding maize for quality and again contains conclusions worthy of attention. Authors representing the Hungarian National Committee for Technical Development (Kralovánszky et al. 1984) emphasize the urgency of this subject for breeders in Hungary and the need to differentiate research according to the type of utilization intended (seed, silage, industrial, human consumption). Of the Hungarian protein requirements 40–45 % originates from maize, so the biological value of the crop is of no slight concern. The use of  $\text{O}_2$  led to a 72–85 % increase in the biological value and, when used as feed, a 12–15 % improvement in the daily weight gain (Szelényi-Galántai; in Kralovánszky 1984). Pásztor and Kovács (1984) report on the inheritance of lysine content and the study of physiological traits of importance from an agronomic position in the case of SC-DC hybrids and the  $\text{O}_2$  hybrids included among the KOTS-2 experiments. Several authors deal with the need to reconcile methods used for the evaluation of quality, though they do not always agree on what method is best (Stehli, Jordánka and Crnobrnja 1984). These authors consider the protein-lysine-tryptophane ratio to be the best indication of nutritional value (the principle behind the CIMMYT method), while others use different principles to determine the nutritional value (e.g. limiting amino acids, essential amino acid index, biological value). The need to increase the quantity of protein and improve its quality continues to be emphasized by foreign researchers (Rotár et al. 1984), who insist that at least one parental pair should have better quality. These authors produced a large number of  $\text{O}_2$  hybrids (1200), 19 of which were entered for state variety trials, where Moldaviai 423- $\text{O}_2$  was given state registration. This hybrid yields 10.4 kg/ha more lysine than the standard, a relative increase of 61.9%. Soviet researchers deal with selection for increased endosperm hardness (using stabilizing genes) and with an improvement in the carotene content. Researchers from Moldavia consider that the nutritional value can best be improved by



reducing the lignine content and improving the N ratios. In zoophysiological experiments the digestibility value was 79–82% (relatively 69–71%) better for conserved corn. Work is also reported on distant hybrids (in Rotár et al. 1984), and the need to utilize further exoplasms (chiefly from Central America) is again emphasized (Hajinov 1972; in Cserbák 1984).

In the *third chapter* several authors mention the importance of broadening the genetic basis of future maize breeding (Tomov 1984). Some inbred lines are not used in any hybrids, while others are found very frequently (e. g. A-632, Mo-17, B-17). Bulgarian researchers consider that 2.5–3.0 t more yield, 12–13% protein, 4–6% lysine and 8–10% fat content can be achieved by breeding, and that hybrids capable of utilizing solar energy more efficiently could also be produced. The amylase, sugar, carotene, vitamin, etc. contents of maize could also be increased. Much of what nature accumulates in maize is still not exploited (Tomov 1984). The negative correlations currently experienced can be overcome through developments in basic research (e.g. genetic engineering). Far-sighted basic research is an essential. Many authors expect results from lines arising through mutations and suggest the existence of "mutational heterosis" (Khristov et al. 1984). Others have studied the pleiotropic effect of the  $O_2$  gene on the zein-carbohydrate ratio and found that the effect of this gene on synthesis deviated in four lines (Rafalski et al. 1984).

The book also contains mathematical evaluations connected with maize research. One such analysis of the economic effectiveness of production (soil, forecrop, cultivation, hybrid, propagation grade, energy, etc. factors) is presented by Petrich et al. (1984). The aim is to forecast the efficiency of production and to measure its effectiveness. The model was set up using data from a substantial number of Yugoslav farms. Ivanovics et al. (1984) report on the physiological and biochemical factors of lodging (mechanical resistance). Stefanovics (1984) recommends a combination of agrotechnical and chemical measures for the weed control of

maize. As a consequence of chemical control, resistant species have evolved (*Setaria* sp., *Cirsium* sp., *Convolvulus* sp., *Echinochloa crus galli*, etc.). The species *Amaranthus* and *Panicum* are now resistant to herbicides. Consequently, chemical treatment should be strictly limited in the future and combined to a greater extent with argotechnical and mechanical measures.

Further literary references are to be found either within or following many of the papers. Both as a whole and in its separate sections, the book provides an excellent review of the major goals and results of the maize research carried out by the CMEA countries within the framework of the KOTS-2 project.

A. KOVÁCS

J. TAMÁSI: *Root location of fruit trees and its agrotechnical consequences* Akadémiai Kiadó, Budapest, 1986.

For a long time scientific research neglected studying the root system of fruit trees.

It was only a couple of decades ago that the fruit growers began giving their attention to the morphological structure of the underground shoot system of fruit trees and the location of roots. In an intensive system of fruit growing the number of trees per hectare may even be multiple, in comparison to the earlier practice. In closely spaced orchards 3–4 years after plantation the root system makes use of the total area available. The tractors carrying out the agrotechnical operations often follow the same track, thus packing the soil in an extreme measure. In the airless soil the roots of fruit trees are unable to fulfil their functions. Changes in the use of rootstocks and the development of soil cultivation methods equally demand a better knowledge of the location of roots and of the factors that modify it.

Tamási has summarized the major results of nearly thirty years of research work in this book. The number and elaborateness of the root exposures done by him are without



parallel elsewhere. The results of his investigation concerning the root systems of fruit species in the temperate zone are of international importance. The English publication of his research results greatly contributes to the appreciation of fruit growing in Hungary.

In 192 pages the book discusses the peculiarities of root location for apple, pear, cherry, sour-cherry, apricot, plum, peach and walnut trees. The examination data are summarized in 61 well-constructed, clear tables. Fine illustrations — 133 in number — complete the work. The pictures clearly show the indefensibility of the so-called drip line theory, which states that the root system of the fruit tree is confined to the area covered by the crown. The examination data also answer the question of whether the extension of the root system of fruit trees is influenced by the type of soil and other characteristics of the plantation, besides the age of the rootstock and the trees.

Along with the examination results the author briefly surveys the methods suitable for studying the root systems of fruit trees. He used the so-called skeleton technique, the most labour intensive but at the same time most valuable method in his work. By this method the soil surface of the spacing of the tree is divided into squares of 1 m<sup>2</sup> each, a unit which forms the basis for the exposure and evaluation of the roots.

Before the evaluation of the examination data we find a comprehensive description of the structure of the root systems of fruit trees, the factors acting on the development of the roots, the periods of root growth, the salt- and frost tolerance of the roots and the agrotechnical factors which influence the formation of the root system.

The sub-chapter discussing the regenerative ability of the root system of fruit trees contains new experimentally proven data which greatly assist the indispensable work of subsoiling and deep fertilization in an expert manner.

The most valuable part of the book is the second chapter which deals with the peculiarities of root location and soil cultivation in

the different fruit species. This chapter is based completely on the author's practical investigations. Data publication of this kind covering a wide range of fruit species is not known to have appeared so far in the relevant literature.

The most important fruit species of the temperate zone is apple. Therefore the root system and its characteristics are dealt with in the fullest detail for the apple tree. The many-sided examination of the root system of apple characterizes the content of the book. Comparative data of the root systems of apple trees of various age with seedling and M4 stocks, respectively, planted in different soil types convincingly show the differences in growth trend between the two rootstocks. At the age of 6 years the diameter of the root system of apple trees with seedling stocks planted in sandy soil is 2.3-times larger than the diameter of the crown, while the root systems of trees of the same age with M 4 stocks have a diameter 2.1-times larger on sand and 1.7-times larger on loam than that of the shoot system.

Pear trees with seedling stocks develop a poorly branching, deep, penetrating root system; at the age of 5 years the extension of the root system of pear trees with seedling stocks is 2.3-times larger than the diameter of the crown. In the case of bearing pear trees the root density within the drip line was found to be 4.3-fold.

Cherry- and sour-cherry trees on *Prunus mahaleb* stocks have well developed, thickly branching root systems. Beside the relatively high sucking power of their root system, the rather good drought tolerance of the sour-cherry trees can be attributed to the well-developed root system. At the age of 23, a sour-cherry tree has 185 meters of roots larger than 2 mm in diameter and these fill a 69 m<sup>2</sup> area. Of this total root length 78.9% is located within the so-called drip line, the area covered by the crown.

The root system of the seedling apricot stock is richly branching; of all apricot stocks it is the one that develops the deepest penetrating geotropic root system. Of the plum stocks the author only examined the



root system of the Myrobalan both before and at bearing age.

Exposing the root system of a walnut tree grown on sandy soil from seed the author found no roots in the upper 0–20 cm cultivated layer. In sandy soil most of the transporting roots of the walnut tree are located at a depth of 41–60 cm. In walnut plantations with sandy soil soil cultivation to a depth of 20–25 cm can be safely carried out.

In the last chapter of the book the author gives instructions as to the depth of soil cultivation with the various fruit species, and the area to be supplied with fertilizers. This may become necessary in bearing orchards in particular, with the view of introducing nutrients or improving the air conditions in the soil. Deeper soil cultivation without damage done to the root system is only possible when the location of the roots of each fruit species in the different types of soil is known.

The bibliography contains 155 original basic publications including 27 references to the author's own works which may supply valuable information to those interested in the subject.

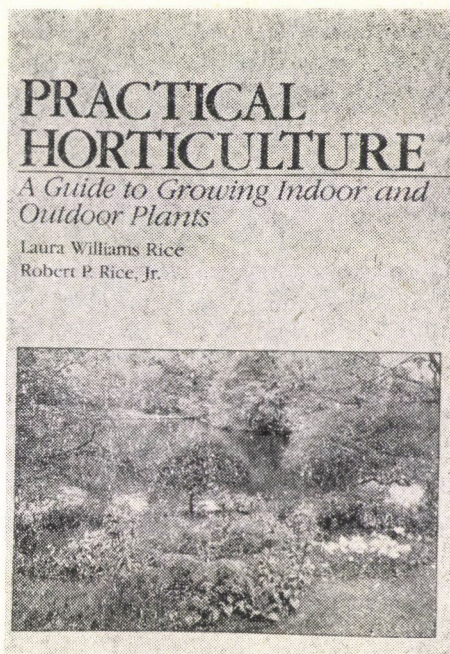
The fine design of the book reflects the editor's exemplary care, and is worthy of the rich content.

Tamási's book published by the Publishing House of the Hungarian Academy of Sciences fills a gap in the relevant international literature. It offers useful information primarily to researchers, teachers and professional growers concerned with fruit growing in the temperate zone.

J. PAPP

LAURA WILLIAMS RICE, ROBERT P. RICE JR.: *Practical Horticulture. Guide to Growing Indoor and Outdoor Plants*. A Riston Book, Prentice Hall, Englewood Cliffs, New Jersey, 1986

This large, finely designed, expertly compiled and richly illustrated book consists of three main parts.



The first part discusses the fundamentals of horticultural production in 76 pages.

In the second part the outdoor plants are described in 226 pages.

The third part deals with the cultivation of indoor plants in 145 pages.

The chapters are completed by a plant-sociology summary, an index of the professional and scientific terms occurring in the text, a list of the authors of the illustrations found in the chapters and a name and subject index.

Some 400 illustrations in the text, and 60 tables render the work easy to use.

At the end of each chapter a list of the relevant literature, along with the names and addresses of the companies and societies dealing with the plants discussed in the chapter are given.

The book supplies scientifically well-founded information about the horticulture of the North-American continent primarily for horticultural students and practising horticulturists. Nevertheless, gardenlovers, hobby gardeners and indoor horticulturists may also find it useful. This work is totally



different from the popular horticultural books, as it teaches on solid scientific bases what to do and how to do it. Yet, its fluent, clear style and language, and the excellent construction make its content easy to comprehend.

The book is a high level, middle grade work.

In the first part the scientific bases of horticultural production are discussed as fully as necessary for the reader to understand the material of the two subsequent parts.

This part deals with the climatic and meteorological conditions, the effects of macro- and microclimate on cultivation, the regions, zones and districts of cultivation in North-America; with the nomenclature, botany and biology of horticultural plants, underlining the biochemical substances which influence the development phases of plants; with the fundamentals of reproduction and propagation, etc.

In the second part the outdoor production of horticultural crops is treated.

Among the environmental conditions of outdoor cultivation great emphasis is laid on questions of soil science and nutrition biology.

A special chapter is devoted to the subjects of field production of vegetables, fruit-growing, outdoor ornamental cultivation. A sub-chapter discusses the fundamentals of landscape-architecture and then sums up the special tasks of a garden designer. Problems of country planning are also touched upon. A special chapter deals with garden-care and plant protection.

In the third part the cultivation and use of indoor plants are treated. The first chapter deals with special questions of indoor ornamental cultivation (e.g. soil and soil mixtures, soil substitutes, water- and nutrient supply, special cultivation methods, etc.).

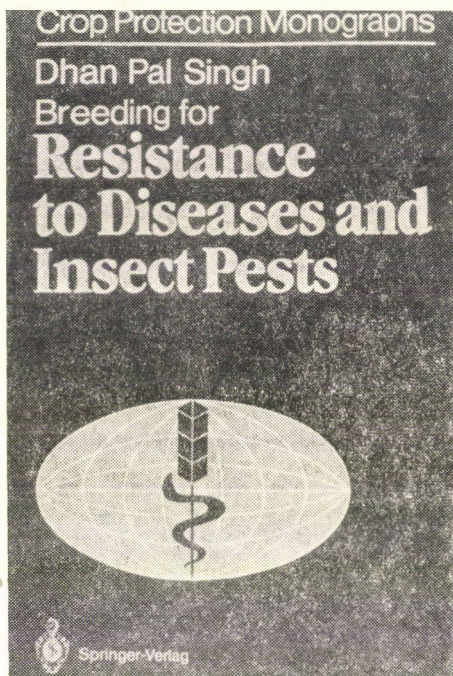
A chapter is devoted to the special cultivation methods of some ornamentals (e.g. Bonsai, Bromelias, Cactuses, Orchideas), as well as to the arrangement of inner spaces, hobby greenhouses and indoor glass cabinets, and to indoor plant protection.

This high level book could be a useful manual for teachers engaged in primary-,

secondary- and higher education, as well as for students, practising horticulturists, and even to lovers of plants with an elementary training.

B. NAGY

DHAN PAL SINGH: *Breeding for Resistance to Diseases and Insect Pests*. The book on Breeding for Resistance to Diseases and Insect Pests is the third volume of Crop Protection Monographs edited by Springer-Verlag, 1987.



The object of publishing the book was to provide insight into the principles of disease and insect-pest resistance and to elaborate the resistance breeding practices with specific examples from as many different crops and parasites as possible. This is partly served by the presentation and evaluation of more than 1200 references.

The 222 page book, including 19 illustrations and 28 Tables, was written by Professor Dhan Pal Singh (Department of Plant Breeding, Govin Ballabh Plant, University of



Agriculture and Technology, Pantnagar, India). The up-to-date knowledge of resistance breeding is presented in 8 chapters.

*Chapter 1* briefly summarizes the value of disease and insect pest resistance, and recapitulates some of the major famines or food losses of crop plants associated with pest and disease epidemics in the past (1940–1980).

A precondition for the development of disease-resistant varieties is that the plant breeder must understand at least the mechanisms of variability in the important plant pathogens, including fungi, bacteria and viruses with which the breeder is confronted in developing resistant cultivars. Equally important is the meaning of host plant resistance. Thus the *second chapter* deals with the variability in plant pathogens as well as the types and mechanisms of resistance.

In 1951 Painter wrote the first book on insect resistance in crop plants, and publicized the fact that the use of resistant crop plant was an ideal way to protect crops against insect pests. The concept, degree, types and genetics of resistance are presented in the *3rd chapter*.

In *four chapter* titled "Genetics of Host-Parasite Interaction" the author describes the biochemical and genetic basis of gene-for-gene and protein-for-protein hypothesis. A scientific response is made to the question "How does the gene-for-gene relationship come into the picture?"

In the case of breeding varieties is resistant to diseases and insect pests, it is imperative to search for sources of resistance i.e. donors from which the resistant gene(s) may be transferred. The *fifth chapter* discusses the importance of germplasm as a natural source of resistance. In addition it also describes the screening techniques.

The conventional methods of breeding for resistance are summarized in cross-pollinated, self-pollinated and vegetatively-propagated crops in *six chapter*, with special regard to the management of vertical and horizontal resistance.

In *seven chapter* the unconventional techniques i.e. plant cell-, pollen-, and protoplast

cultures, mutant cell selection, genetic engineering and their possibilities are summarized.

This book can be recommended for use as an advanced text and as a reference book for plant pathologists, entomologists, and plant breeders engaged in developing varieties resistant to harmful parasites.

L. E. HESZKY

R. PETERS and M. TRENDELENBURG (eds): *Nucleocytoplasmic Transport*. Springer-Verlag, Berlin, Heidelberg, New-York 1986. (300 p., 115 figs and 12 tables)

The eukaryotic cell contains numerous compartments separated by membranes. These membranes play a fundamental role in separating the specialized parts of the cell and at the same time they regulate the interconnections between these parts.

One of the important differences between the prokaryotic and the eukaryotic cell is that the nucleus of the latter is separated by a double membrane from the cytoplasm. The transport of materials between the two cell parts is made possible by the pores of the nuclear membrane through which the smaller molecules easily diffuse, while the larger molecules such as proteins and nucleic acids are transported by the participation of a special pore structure.

The molecular details of the relations between cell nucleus and cytoplasm have only recently received close examination, largely with the up-to-date methods of genetics. The opportunity to encompass the present range of knowledge on the subject was offered by a workshop on the nucleocytoplasmic transport held at the German Cancer Research Center. The proceedings of this workshop were published in 1986 by the Springer-Verlag.

The book contains the text of 24 lectures grouped in 4 parts. After the introduction and a historical survey (J. Brachet, Brussels) 8 publications deal with special methods for studying the membranes, 4 with the structure



of nuclear pores, 9 with the transport of proteins and 2 with that of ribonucleic acids.

Of the lectures on the techniques of examination those by M. Engelhardt et al. (Munich) discuss spectroscopic and special microscopic methods used to measure the elastic properties of membranes; P. B. Garland and J. J. Birmingham (Dundee) describe an optical method of measuring the displacement of membrane proteins; G. A. Kraft and co-workers (Syracuse) acquaint the reader with a new method for measuring the diffusion of a fluorescent markers; C. M. Feldherr (Gainesville) shows how to use colloidal gold tracers to study the nuclear transport; G. Neuhaus and H. G. Schweiger (Heidelberg) demonstrate the suitability of the *Acetabularia* model for studying the relations between nucleus and cytoplasm; N. Riedel et al. (Frankfurt am Main) suggest the use of vesicles prepared from the nuclear membrane as a model system; J. E. Edstrom (Lund) examines the transport of RNA molecules to the cytoplasm with a microdissection; technique finally, M. Trendelenburg et al. (Heidelberg) discuss the possibilities of studying the chromatic organization nuclear by means of electron and light microscopy.

The structural model of the nuclear pore complex is described by R. A. Milligan (Stanford); G. G. Maul and G. Schatten (Heidelberg) discuss the characteristics of lamins, the main protein components of the nuclear membrane; Chr. Dreyer et al. (Tübingen) study the protein uptake by the nucleus with the aid of monoclonal antibodies.

In the series of lectures on the nucleocytoplasmic transport of proteins C. Dingwall et al. (Cambridge) treat a selective mechanism promoting the transport of proteins into the nucleus; B. Schulz and R. Peters (Frankfurt) describe a specific signal sequence for the nucleoplasmin; similar specific sequence determines the location of proteins within the nucleus, according to the results of experiments carried out with SV 40 virus proteins by B. L. Roberts et al. (London, Berkeley) and W. Deppert and M. Staufenbiel (Ulm) a. Colman and J. Davey (Coventry) used influ-

enza virus, while B. D. Richter and N. C. Jones (London) applied adenovirus as the model for studying the nuclear accumulation. A localization signal sequence was of proteins also demonstrated on yeast nucleoproteins by M. N. Hall (San Francisco). The molecular mechanism of hormone action was dealt with by A. Dierich et al. (Strasbourg) and U. Gehring (Heidelberg).

At last, the importance of the RNA-protein interaction in the intracellular transport was discussed by I. W. Mattaj (Basel), and the role of ribonucleoprotein particles in RNA processing by U. Skoglund and his team (Stockholm).

All in all, it can be concluded that while most contributions analyse various details and present recent data of particular interest mainly for specialist working in the respective line, some reports attempt to draw conclusions and outline general models interesting for everyone attracted by cell biology, thus giving insights not only into the details of scientific research but also into the wonderful mechanism of function of the living cell.

This book with its high scientific level and defined scope of subject can be mainly recommended to researchers and specialists.

T. DEÁK

*Systematische Botanik*, Herausgeber: Dahlgren, G. Unter Mitarbeit von: Björkquist, I.; Dahlgren, R.; Nilsson, Ö.; Runemark, H.; Snogerup, S.; Weimarck, G., VIII. 259 p., 436 figs. Berlin-Heidelberg-New York-London-Paris-Tokyo: Springer-Verlag 1987.

The "Introduction" explains the concepts of taxon, population and species. The number of all species of the flora cannot even approximately be assessed, since the species numbers of large areas in the world are still unknown. Later with each group of plant an approximative value indicates the number of species known to belong to the group. It is interesting that the material in worldwide botanical collections runs to more than 100 million herbarium specimens of seed plants



alone. Taxonomic units and scientific systematization, are mentioned, with reference to the rules of the "Internationalen Code der Botanischen Nomenclatur". The roles of palaeobotany, morphology, chemistry, cytology and of different reproductive methods (sexual, asexual, vegetative) and of the alternation of generations in plant systematization are discussed. A clear survey is given of production economy, energy and nutrient consumption (autotrophy, heterotrophy) as taken into consideration from the standpoint of systematization.

The description of the system proper begins with the chapter "Gliederung der Organismenwelt", showing the structure and the cell components of the prokaryotic and the eukaryotic cells. With the new classification of living organisms (*Monera*, *Fungi*, *Plantae*, *Animalia*) the author finds it difficult to consider the fungi a separate uniform group.

In the general characterization of prokaryotes, the book indicates that the viruses with their replication mechanisms differ from all the other living organisms. Accordingly it divides the prokaryotes into *Archaeobacteriophytes* (Archaeobakterien), *Eubacteriophytes* (Eubakterien) and *Cyanophytes* (Blaualgen). Morphological features, conditions of division and occurrence, and major representatives of the group are described, even for the *Cyanophytes*, an approximative number of species (1200) is given.

The chapter on *Eucaryotes* begins with a general characterization of Algae: from the morphological differentiation, cell organization, sexual and asexual reproduction, life cycle and nutrition, to the technical production and systematization of algae. They are placed in 9 phyla and 2 sub-phyla:

- (1) *Chlorophytes* (c. 8000 species)
- (2) *Euglenophytes* (c. 400 species)
- (3) *Pyrrophytes* (c. 1000 species)
- (4) *Xanthophytes* (c. 360 species)
- (5) *Chrysophytes* (c. 325 species)
- (6) *Bacillariophytes* (c. 6000 species)
- (7) *Phaeophytes* (c. 2000 species)
- (8) *Rhodophytes* (c. 1500 species)

- (9) *Mycotes* (fungi) (c. 60.000 species/discussed on 25 pages with 2 sub-phyla: *Myxomycotines* (c. 600 species) and *Eumycotines* (c. 60.000 species))

The *Cormophytes* are reintroduced by a general characterization. Most of their representatives are phototropic; saprophytes and parasites are few among them. The cell wall consists of cellulose. The apical meristem of shoot and root is considered by the author to be an important characteristic of the *Cormophytes*.

The phylum *Bryophyta* (c. 20.000 species) is a well-defined group of the simplest *Cormophytes*. After a phylogenetic survey, the characteristics of gametophytes and sporophytes in the classes *Marchantitae*, *Sphagnatae* and *Bryae* are described. The phylum *Pteridophyta* (c. 10.000 species) is divided into 4 characteristic sub-phyla (*Rhyniophyta*, *Lycopodiophytina*, *Equisetophytina*, and *Polypodiophytina*).

The phylum *Spermatophyta* includes the largest number of species (c. 220.000). These most diversely differentiated plants are peculiarized by the way they form their seeds. According to whether the seed stands free or is enclosed by the fruit wall, they are divided in 2 sub-phyla: *Gymnospermae* and *Angiospermae*. Of the *Lygnopteridatae*, *Cycadatae* and *Pinatae* classes of *Gymnospermae*, a brief survey is given.

With the *Angiospermae* a special morphological chapter deals with germination, germ-plant, root, shoot, leaf, modified and special shoots, flower, seed, fruit and inflorescences. The chapter "Embryology" describes the development of embryo, pollen and embryo sac in *Angiosperms*, the formation of pollen tetrads, the pollen germination and fertilization, the development of endosperm and embryo, the seed development and the apomixis.

In the chapter "System" the systematic importance of the different (palinological, embryological, anatomical, chemical, cytological, palaeobotanical) characteristics of seed plants is demonstrated. By representing Dalhgren's phylogenetic tree for *Angio-*



*spermae*, the book gives the basic principles of classifying the seed plants.

The species number of dicotyledons (*Dicotyledonae*) is about 165.000. Its subclasses are: *Magnoliidae* with about 35 families and more than 10.000 species; *Hamamelidae* with a few families and some 3000 species altogether; *Caryophyllidae* with 14 families and 11.000 or so species; *Dilleniidea* with nearly 24.000 species in 70 families; *Rosidae* with about 60.000 species in some 100 families (lignified and herbaceous plants with a great variety of forms); *Asteridae* with about 56.000 species belonging to 55 families; and *Alismatidae* with only 500 species in 10–12 families.

The subclasses of *Monocotyledons* are: *Commelinidae* with about 19.000 species in 15 families; *Liliaceae* with c. 30.000 species from 25–35 families. Each subclass and the major families are characterized in a few words. The species number of monocotyledons is estimated at 55.000 altogether.

The chapter on "Cultivated plants" is divided into these sub-chapters: history and growing of cultivated plants; plants as nutrients; plants as spices, stimulants and medicines; plants producing fibre, rubber and timber.

The chapter on "Reproduction" biology deals with the biology of pollination and distribution.

In the chapter "Life form" the duration of life, the process of lignification, the role of overwintering organs, and the correlation between life form and water regime are discussed.

The chapters in the section of "Plant geography" deal with the area types and with the effect of postglacial changes on plants, and describes the flora regions of the Earth (Holarctic, Palearctic, Neotropis, Australis and Capensis) and of the sub-regions of Europe (arctic-, boreal-, Central European-, atlantic-, mediterranean-, pontic-, and turanic).

The book is completed by a rich "Bibliography", a scientific name- and subject index, and a geological time table which informs the reader of the appearance of the major

groups of plants over millions of years.

The richly illustrated book offers a clear view of both the flora and the taxonomic role of the related branches of science. It can be used with equal profit by teachers and students of universities and colleges, and may be useful in the postgraduate education of those working in any special field of botany.

I. MÁTHÉ

A. BRAITHWAITE and F. J. SMITH: *Chromatographic Methods* 403 p. 165 figures, 28 tables. ed.: Chapman and Hall, London, New York, 1985.

For several decades analytical and preparative variations of the chromatographic methods have become indispensable means of examining organic and inorganic matters.

The book of "Chromatographic Methods" is a thoroughly revised and up-dated work in comparison to the existing handbooks on this subject. Its nine chapters present the theory and practice of chromatography, and offer detailed information about the practical methods employed in industry and research.

The first chapter surveys the history of the chromatographic methods and classifies the techniques used in current practice.

The theoretical considerations concerning the various methods, possibilities of influencing the factors of retention, fundamentals of the capacity of chromatographic columns, of the resolving power, of the shifting of bands and efficiency of separation are discussed in a special chapter.

Within the planar methods of chromatography the book deals with thin layer chromatography, paper chromatography and electroforesis. In the chapter that encompasses the adsorption chromatographic methods, the chromatographic techniques based on the principle of adsorption, the fundamentals of the static- and dynamic phase, and relations describing the efficiency of separation are established.

Another chapter discusses the methods of gas chromatography and the high capacity



techniques of liquid chromatography, their principles and the possibilities of carrying them into effect — including the critical points of instrumentation.

Illustrations and examples assist in processing the data of chromatographic examinations.

The last chapter contains a series of experimental models which represent various methods used in practice.

Although the size of the book does not allow completeness, no area of any practical importance is ignored. Chemists and biochemists of laboratories applying the above techniques of separation, as well as university students and technicians who want to become acquainted with the subject, will certainly use this book profitably.

ÉVA SÁRDY

#### *Drip irrigation congress, California, 1985.*

The 3rd International Congress on drip irrigation was organized in Fresno, California, from 18 to 21 November 1985. The previous congress was held in San Diego. That these took place twice in succession in California is certainly related to the fact that, despite its rather dry climate, this region is one of the most intensive agricultural areas of the world, with horticulture as the main branch of cultivation due to the various irrigation methods. Owing to the scanty supplies of water, drip irrigation with its economical efficiency has become a widespread practice. The industry which manufactures the equipment displayed its supply of goods at the congress exhibition.

Most of the lectures — 85 in number — were given by the representatives of the organizing countries. Israel was represented by 11, and Australia by 5 lectures. Hungary took part in the congress with 3 lectures.

The subject matters of the lectures were published in two volumes. The *first volume* concerned the following subjects: the perspectives on irrigation, study of (economic consideration) the economic conditions, clog-

ing and water treatment for irrigation, drip experiences in the United States, international experiences, crop production and response to irrigation, field evaluation of drip land utilization irrigation, product testing and evaluation, fertigation and drip irrigation design.

The *second volume* also covers some interesting subjects: chemigation, insect and weed control, aspects of chemization, nursery cultivation, comparison of drip irrigation to other irrigation methods, drip systems and equipment, irrigation with saline water, root-system development soil and infiltration, subsurface drip irrigation.

Owing to the large number of participants from the United States a full description was offered of the distribution and development of dripping irrigation there. We find this important, primarily because most of the statements agree with the Hungarian experiences.

#### *The role of drip irrigation*

Drip irrigation — like other methods of irrigation — is a technique of water supply which serves to maintain plant life and develop plant products. This method of irrigation is highly efficient for supplying the plant with water. This relatively new solution renders possible a much more accurate utilization of water. At the same time it requires a greater knowledge of how to operate the irrigation systems so as to increase production.

#### *General experiences of dripping irrigation*

For the last fifteen years much experience has been gained in the drip irrigation, and a great increase both in scientific results and everyday practice has been achieved. In 1985 some 600,000 ha were irrigated by this method.

This method of irrigation — like the earlier ones — was initially used in the arid zone, but later was also introduced in the humid zone in order to make up for water deficiencies during dry periods.



Earlier scientific experiments carried out with various crops led to some exact results. In particular, the correlations concerning the opening of the water-supplying elements and the filtering of water were determined. The reliability of the newly-developed water chargers and water filters has increased, their work has become more precise, and therefore they have found an ever wider experimental and practical application.

For areas in which the soil is rich in nutrients, the temperature is high and sufficient water is available, the conditions of efficient irrigation farming are given. The irrigation farming that produces increasing yields has become an active participant in the competition for the existing water supplies. The continually decreasing water supplies and the rising energy prices urge the users to find solutions whereby the use of water and energy can be reduced. The improvements in water management — including the economical use of water supplies and the development of irrigation technologies — greatly help to change or eliminate the less efficient irrigation systems.

The earlier and, partly, the present sprinkler irrigation systems (hand-transferred-, hauled- and self-propelled branch-pipes) have some typical disadvantages:

- high rate of water supply with the possible consequence of surface running-off,
- high labour demand (with hand transfer),
- high maintenance- and energy prices.

At the same time, there was a possibility of utilizing and developing further the advantages of these systems by a new method of irrigation. The shortcomings can be reduced or eliminated with a properly designed and constructed drip irrigation system.

At the beginning the Californian growers confronted two problems:

- (1) clogging of emitters
- (2) changes in the water supply of plantations from the earlier surface- and sprinkler irrigation methods wetting large volumes of

soil to a solution when only a part of the soil is moistened.

To prevent the emitters from clogging they suggested the following methods:

- to establish water reservoirs as sedimentation basins (for settling floating impurities which may block the filters),
- to apply chemical solutions for reducing the growth of algae and aquatic plants,
- to choose emitters with larger diameters,
- to overplan the capacity of the equipment by 50%.

Those in the vanguard of agricultural production were of the opinion that these planning considerations and the proper operation of the systems would render the irrigation of any crops possible.

Soon after the establishment of the first irrigation plants it became clear that the regular and thorough flushing of the system was indispensable for the drip irrigation equipment to work without clogging. The efficient cleaning of the system included a chemical treatment against algae present in the pipes.

The first problem arising on the operation of the irrigation plant was the reservation with which the operating staff accepted the new solution. They thought that the drip irrigation was not able to supply the plants with sufficient water. The change was received with doubt not only by the farms but also by the consultants.

Though at the beginning some failure did occur, the attempt, as a whole yielded positive results, and this convinced the operators of the irrigation plants. The as the method spread, the operators became its supporters. When they accepted the conception and became aware of its yield-increasing effect, they produced unexpected results.

The new laterals, though, moistened, smaller volumes of soil, which could be compensated by an increase in the frequency of irrigation. The root-systems of both citrus and grape-vine proved to make better use of the quantities of water available in a reduced volume of soil.



The initial fear — of changing over from the traditional to the drip irrigation of developed woody plants — appeared to be unfounded. For the almond the replacement of sprinkler by drip irrigation was no as efficient as for the citrus and the grapes. The fully developed almond-trees did not respond satisfactorily to drip irrigation, therefore minisprinklers were subsequently used the moistened larger volumes of soil and roots which put an end to the earlier anomalies.

#### *Operation of the first drip systems*

The initial aim of drip irrigation (as mentioned before) was the economical use of water and energy and the reduction of the labour cost. In California — unlike Australia — the first irrigation plants were established with highly refined pump- and control systems. The pumps, wells and drip irrigation facilities were under automatic control. The water level in the reservoir was automatically controlled and the reservoir was replaced according to the water demand of the irrigation plot. The purpose of automatization was to decrease the labour demand. With an increase in the working hours the number of maintenance hours turned out to exceed by far the originally expected demand. While the amount of labour earlier used for irrigation work decreased, the number of hours spent in maintaining the technical equipment increased. (Part of the labour cost savings was used up by the technical staff. The application of traditional planning parameters which increased the initial sums of investment had a cost-increasing effect.)

They were:

- (1) The dripping units of 8 l/hour water discharge increased the volume of water to be delivered; that is, pipes of larger diameter had to be used.
- (2) The traditional water norm per plant likewise demanded a larger volume of water delivery.

The parameters applied resulted in a higher hydraulic capacity and higher investment costs.

#### *Operation of recently developed dripping systems*

The twenty years that have passed since the beginning provided sufficient experiences in the field of planning, construction, operation and maintenance. Most farms have found the methods and means best adaptable to the crops grown. The initial clogging of water chargers has been successfully minimized by filtering and by proper maintenance of the equipment. The latter consists of a regular flushing out of pipes and removal with chemical substances of microorganisms (algae) present in the system.

As a result of the careful operation, about one third of the facilities worked perfectly even after 15 years. The development work resulted in new facilities every years. As a consequence all systems worked well and hardly any decrease in output occurred. Accordingly, a highly profitable management was attained. From the yearly alterations, those doing the development work gained considerable experiences. By proper cultivation and maintenance, drip units of lower water discharge and smaller diameter can even be operated without clogging.

Next it was realized that the operation of the system had to be simple enough to maintain the safe discharge of water and involve the least possibility of failure. As a result most of the filter- and pump systems today are hand operated. The lower capacity irrigation systems have a longer life and still are able to moisten sufficient volumes of soil. By using pipes of smaller diameter the investment cost can be reduced.

An important stage of development came when the daily water requirement for almost every woody plant was determined. The lower capacity equipment, the hand control and the properly determined daily water demand made it possible to reduce the costs by one-third compared to the initial period. Further improvements, such as the correctly chosen material of filters and emitters, greatly contributed to the reduction of production costs. The knowledge acquired enables us to take into consideration the corre-



lations of soil, plant and water in planning, whereby the best approach to irrigation systems of cheap operation can be achieved.

*Summarizing:* those undertaking development work performed tiresome and costly tasks in the course of learning. The initial planning, equipping, operating and maintaining factors and values gradually changed on the basis of the experiences obtained.

Those taking part in the development work have arrived at the conclusion that a proper way of planning, equipping, maintenance and operation is necessary.

This includes:

- a system built up with an engineer's accuracy, with the hydraulic and economic factors taken into consideration.

- installation work done by skilled mechanics,

- efficiently working flush system,

- properly trained operators.

If these important factors are taken into consideration there is no doubt that the drip method of irrigation will be chosen in arid and semiarid regions where the water supply is the main limiting factors of crop production.

F. LIGETVÁRI

*Information on maize* (Információknjű Bjűl-leteny po Kukuruze). Koordinacijnű Centr SZEVI po probleme KOC-2. Naucsno-izledovatylszkij Inst. Sz/H. VAN Martonvűsűr No. 5. 1986.

This publication of 182 pages was compiled by the Agricultural Research Institute of the Hungarian Academy of Sciences. It primarily describes the results and directions of research carried on in the socialist countries in the field of breeding maize for quality of its components. The improvement of the quality of maize grain is the subject of 10 papers (Part I.), and the increase of seed-grain production by 4 papers (Part II.): the papers include well-constructed tables and figures. Many authors provide bibliographies also.

Several papers outline the situation, timeliness and perspectives of breeding maize for quality (K. Zima, A. Normov, K. Jordanova et al., K. Sztasztni et al., A. Rotűr et al. and G. Karaivonaov et al.). Authors from the Soviet Union, Bulgarian People's Republic and Federal Republic of Yugoslavia report on qualified O<sub>2</sub> hybrids (e.g. Kr 456 L, Kr 461 L, Kr 333 L; Zima et al.). In other socialist countries the production of hybrids of commercial value is considered possible only with early maturing hybrids (J. Naether et al. in the German Democratic Republic; some authors in the Hungarian People's Republic; and several other socialist countries); they think that the higher value of components and the agronomical characters (e.g. yield) are difficult to bring to accord, particularly in the case of the medium or late — that is, mostly the high yielding — maize varieties. Besides the improvement of the seed-grain, several authors deal with the nutritive value of the green crop and of the CCM fractions (Naether et al., Roter et al. and Sztasztni et al.). Moreover, some of them consider the latter more favourable for more northerly regions. The Hungarian researchers hold a different opinion of this.

The possibility of producing hybrids of commercial value is judged variously in the socialist countries; yet they agree upon the timeliness of the subject.

A number of authors deal with other important agronomical characteristics (dry matter, plant/ear, plant/grain weight, height of ear, length of ear, weight of ear, ear/grain, ecological adaptability, ecological stability, resistance etc.). These investigations concern the most fundamental questions of the scope of this subject.

The O<sub>2</sub>-endosperm mutant is generally used, and is regarded as a landmark in breeding maize for quality of components (K. Zima et al.), which has opened a new phase in breeding for protein content. Earlier the O<sub>2</sub> gene generally was introduced in lines, BC (back-cross) generations were produced combined with inbreeding; then, with them, hybrids were developed. Today increased emphasis is laid on selection for genes modify-



ing the hardness of the endosperm (Soviet Union, Bulgarian People's Republic and other countries), on synthetic populations produced by making use of matutations for mutative hybrid breeding (Bulgarian People's Republic), and tissue culture techniques were even considered as a solution (Bulgarian People's Republic).

The socialist countries try to bring their breeding methods into accord. The use of the proper standard arises as a methodical question. This applies to the uniformity of the chemical analysis methods, whereby the evaluability of the examinations and the comparability of the results of field trials can be ensured. This depends on the means available in the individual countries and situations.

It would be useful to establish composition parameters smaller in number than the present ones but expressing clearly the nutritive value (e.g. lysin/protein, triptohane/protein ratio). In Hungary the evaluation of the nutritive value is based at present on the dry matter-, protein-, limiting amino acid (lysine, triptophane, methionine) content, essential amino acid index and biological value, and occasionally on feeding tests. Investigations connected with the examination methods are reported first of all from the Bulgarian People's Republic, and then from other countries (K. Sopova and K. Jordanova, G. Karaivanov et al., O. Fadajeva et al. and Roter et al.).

Genetic basic research connected with breeding maize for protein content (diallele analysis, inheritance of polygenic characters) is also carried out by a number of authors (Zieger, G., Karaivanov et al. and N. Tomov et al.). Research is extended to cover further components (lignin, fibre), or biochemical examinations connected with other subjects are made (nitrate reductase activity — ANP; O. Fagyejeva et al.).

Since 1985 in KOC<sub>2</sub> experiments some hybrids known to have high lysine contents and many prospective hybrids have been examined in the socialist countries. Registered hybrids are reported first of all from the Soviet Union and the Federal Republic of

Jugoslavia; eight From the latter country and from the Soviet Union new registered hybrids (kr 456 L, KR 461 L, Kr 333 L) — in Hungary there were registered hybrids earlier than in the mentioned countries, though. The hybrid jointly turned out by the German Democratic Republic and the Hungarian People's Republic was qualified in the GDR. (J. Naether et al.). In Hungary the qualification seems to be stricter, that is why there are not registered hybrids here. The high lysine content hybrids must not fall behind the normal maize in other agronomical characteristics. The authors describe several hybrids known to have high lysine contents which are not inferior in agronomical characteristics either (Federal Republic of Jugoslavia). Their introduction in commercial production is still of moderate extent.

Researchers in the Bulgarian People's Republic use synthetic populations — including mutations — to produce a basic material of high lysine content. From this basic material they carry out recurrent selections. The population is homozygous as regards the O<sub>2</sub> gene. The synthetic population includes internationally known good inbred lines and, lines containing modifier genes. The selection for a high lysine content material is made with 8–16 synthic lines and a convergent breeding method. Until 1990 the material will be propagated, and the member countries will also receive from it. The method is expected to result in a higher yield and an ecologically stable, well adaptable hybrid (K. Jordanova et al.). In the cooperation other socialist countries take part too. In the Bulgarian People's Republic the method is considered to be a turning-point in breeding maize for composition value. In the same country tissue culture (embryo culture) experiments are also conducted.

Tomov et al. study the effect of ecological conditions on the agronomical and biological properties (phenological features, yield structure, correlative characteristics) of lines and hybrids, since the socialist countries differ in cultivation — and climatic conditions (relative humidity, day-length, temperature, spectrum composition, etc.). Genetic adap-



tability is examined primarily in relation to ear and ear/grain number both for lines and hybrids. As a result of these studies the authors emphasize the close relationship between genotype and environment.

K. Sopova and K. Jordanova recount their experiences gained by comparing various chemical analysis methods. They point out that the different results (e.g. with protein- and lysine content) may be due to differences in the methods of analysis and the conditions of the given country (research site). They stress the importance of uniform data (the protein should be given in absolute dry matter, the amino acid as percent of protein).

Researchers in the Soviet Union pay increasing attention to maize mutants carrying the  $O_2$  gene which modifies the seed hardness. They have several new hybrids of commercial value and high lysine content carrying  $O_2$  gene. The Kr 333 L. exceeds the Kr 303 which is the standard hybrid of normal endosperm both in yield and feeding value; lysine production (kg/ha lysine) was 40–48% more and its triptophane production 40–60% more. Its stalk strength was not inferior either (Zima et al.). On feeding 45–50% savings were attained with it. It is the grain structure of the maize carrying the modifier gene that has changed together with its biological and chemical properties. The authors emphasize that in the work of breeding maize for high lysine content the lines and hybrids have to be considered as a complex. They report on new high lysine content combinations too (Kr 456 L, Kr 461). The results deserve attention if only because they suggest the possibility of selecting for commercial and quality parameters at a time. The results are influenced by the standard used as well.

Several researchers in the Soviet Union study the biochemical characteristics of the double recessive lines. In breeding maize for quality, a distinction is made between breeding for lysine- and protein content and improving the quality of the green fodder (Rotár et al.). They study the  $O_2$ ,  $bm_1$  and  $bm_3$  mutants and their hybrids, and examine the

lysine and lignin too. They have found that the double recessive lines are characterized by more lysine and less lignin. The authors think that the joint improvement of the components and agronomical characteristics of maize is possible, but this requires and appropriate genetic basic research. On the basis of the results of their investigations basic materials suitable for improving the nutritive value of grain and silage can be produced.

Others (O. J. Fagyajeva et al.) study the physiological characteristics relevant to the production of high protein content maize (biochemistry of processes of assimilation, transportation, storage). They examined the inheritance of the chlorophyll content of leaf surface and activity of nitrate reductase in 27 hybrids. The aim was to lay down the physiological bases of breeding for high protein content. The same authors studied the relationship between grain and green parts (ratio of leaf N/grain N; N index) (Fagyajev et al.).

Of the other components the lipid soluble vitamin content (A-provitamin, E-vitamin, of yellowgrained maizes, as materials increasing the daily weight gain, and the carotene content were subjected to examination. The heterosis of carotene was previously demonstrated (Mihajlovics 1971). The inheritance of these characters was earlier studied by others (Quackenbush) and found homodynamic. Some of the hybrids have now higher carotene contents (Moldáviai 349 K). With this they laid the genetic-physiological foundations of quality breeding for a given species.

This subject is dealt with in the German Democratic Republic (Naether et al.). They report positive results in breeding early maizes for components and improving them at the same time as green fodders, and they describe feeding experiments. In early maize breeding they cooperate with Hungarian researchers (Martonvásár), and carry on CCM (Corn Cob Mix) development too.

The conditions of the GDR make particularly reasonable the improvement of the nutritive value of green fodders (the development of the CCM method), and in this the  $O_2$



mutant is also used ( $O_2$ -CCM). In the GDR endeavours are made to distribute the  $O_2$  hybrids on a farm-scale; at present three prospective varieties are included in a variety trial managed by the state. According to their expectations this work may result in 15% savings of protein in the GDR.

G. Zieger tells of the diallele crossing of early inbred lines. His experiments were carried out in three locations with three plant densities, ten lines and their hybrids and seven testers. The experimental material included hard maize and dent-corn too. The yield and other properties (lodging, wet%, plant/ear, etc.) greatly depended on the conditions (site, plant number, year). Between vegetation period and yield the correlation was negative ( $r = -0.72$ ), while between yield and standability it was positive ( $r = +0.64$ ). The dary matter content showed an additive gene effect.

Several authors from the Federal Republic of Yugoslavia (K. Stasztni et al.) report favourable results of experiments (aimed e.g. at increasing the fat content); investigations with silage maize or other ways of using maize (non-dray grain) are carried on in Hungary as well.

Of maizes with high lysine content, eight have been registered so far. Although their yield is lower than the standard, the water discharge of grain slower, and they are more susceptible to diseases. Yet, some of them are not inferior to maizes with normal endosperm and their components are of higher value. They have not spread in commercial production since the conditions of Yugoslavia are more favourable for soya cultivation (the question is of higher relevance where the cultivation of soya is unreliable). Beside the lysine content, the protein content is also a goal of breeding. For the characterization of the nutritive value some authors consider the lysine/protein- or triptophane/protein ratio also important. They examine the nutritive value of silage, wel crushed grain and CCM. The lysine production of the high lysine hybrids was 48.4% more in 1974–1983). With grain maizes they also carry out selection for the hardseed  $O_2$ . As for fat content the aim

is to increase it from 4% to 8%. Selection is made for amlopectine ratio too (K. Stasztni et al.).

Intensive work is done in the subject at the research sites of Hungary too (Martonvásár, Szeged, GKT, universities and other institutions), occasionally in bi- or multilateral cooperation. Hungary is carrying out the coordination of these investigations Agricultural Research Institute of the Hungarian Academy of Sciences, Martovásár).

On the basis of these papers it can be established that, despite the existing problems in plant breeding, the COMECON countries carry on a many-sided work concerning the improvement of the components of maize, and over several years have made some definite progress.

Part II. of the publication (4 papers) deals with some theoretical and practical questions of maize breeding and seed production. The investigations are aimed at increasing the yield of grain.

In Hungary hybrid grain processing is carried on. The processing of the BEMA TC 211 hybrid was examined from the point of view of how the individual phases of processing affected the quality of grain. In three years the yield from three million ha is processed in this way. During the different phases the quality of grain may be reduced, and when accumulated this may cause differences in the end product. The deterioration of the quality of grain during drying and storage is greater than it is in the course of processing with the Hungarian technology. The shape of grain influences the quality of the hybrid grain after processing. The quality may be reduced in the course of processing, but the proper way of processing may even improve the quality of grain produced under less favourable conditions (Barla Szabó G. et al.).

The relationship between simultaneous flowering and grain quality was also studied. Simultaneous flowering was found to be uncertain with the SC-hybrids. The maturing of the SC-hybrids is also more differentiated. The influence on grain quality of a paternal line flowering late was also examined. Late flowering was found to affect the quality of



grain, and the effect of a 60% fertilization was even felt in the germination percentage, though there were differences depending on the hybrid (Szundy T.).

Researchers from Bulgaria describe their experiences with seed production. They deal with the external and internal factors of the maintenance of grain density during propagation (P. Preszolszkaja). The 12 : 4 ratio of maternal and paternal rows are considered optimum from the point of view of increasing the grain production. Emphasis is laid on the role of genetic sterility, the removal and silaging of paternal rows, and the mechanization of harvesting. The cultivation of hybrids with different ripening times is considered advantageous. P. Hrisztova and K. Hrisztov deal with SC-hybrids modified by mutation. The classical lines and those obtained by mutation were successfully combined. Experiments are conducted on crossing mutant  $\times$  mutant lines. The hybrids thus produced were examined under different ecological conditions (in 5 places, 2 with irrigation, 3 without). With such hybrids (KNM 530, KN 614, KNM 614, etc.) a 26% yield difference was achieved.

As seen from the reports in the socialist countries one of the most timely questions of maize breeding — the improvement of quality — is studied from many sides in the framework of the COMECON-KOC<sub>2</sub> project. The publication supplies useful information first of all on the work of breeding maize for components (Part I.), and on the improvement of the quality of seeds for those interested in the subject.

A. Kovács

L. VAJNA (ed.): 1987. *Fungal pathogens of plants*. 1-303. (68 figures, 230 photographs). Mezőgazdasági Kiadó, Budapest.

The book subtitled "Questions of chemical and biological control" contains four chapters, which are:

(1) Characterization of the fungal pathogens of plants.

(2) Relationship between the pathogenic fungus and its host plant.

(3) Chemical control.

(4) Biological control.

The authors of the book (B. Barna, Maja Gasztonyi, Hornok, Gy. Josepovits, Á. Szécsi, L. Vajna, J. Vörös) and its editor (L. Vajna) summarized the latest results of research in Hungary and abroad in the above subjects. The bibliography at the end of each chapter renders a more detailed study of the subject possible.

The readers of the book must possess a mycological knowledge — in a broader sense a knowledge of plant protection —, if they are to make use of the contents.

The chapter "Characterization of the fungal pathogens of plants" includes the electron microscope-, biochemistry- and serology relations of systematization. They are important for the chemical and biological control too.

In the chapter "Relationship between the pathogenic fungus and its host plant" a survey is given of the extensive research on host-parasite relation and disease resistance, of the results attained and the direction of further investigations.

The chapter "Chemical control" deals with the fungitoxic substances and their active ingredients of the fungi and their host plants. Emphasis is laid on the fungicide resistance, an important problem at present. Further, the authors touch upon the connection between control with fungitoxic substances and environment protection, and examine its contradictions. It is obvious that the importance of this question and the literature on it will increase in the future.

The chapter "Biological control" summarizes the present knowledge of the subject and the results realized in practice. Furthermore, the authors outline all those possibilities which later might play some role in an integrated control.

All in all, the book provides a useful theoretical basis for those engaged in education and research in the field of plant protection.

M. GLITS



*Integrated Pest Management* (Eds: A. J. BURN, T. H. COAKER and P. C. JEPSON Academic Press, London, 1987. pp. 474.

The integrated pest management (Henceforth abbreviated IPM) is not a new idea, but since its birth it has been interpreted in many different ways.

According to the simplest formulation it is a combination of biological, chemical and cultural control techniques.

The book provides a full explanation of what the IPM is in practice, what its components are and how important is the role played by it in reducing the pesticide stress of our environment and preserving the resources of nature. It calls attention to all harmful consequences of the increasing use of insecticides.

The fourteen chapters of the book were written by fifteen authors, as well as the three editors.

In the *first chapter* M. E. Cammel and M. J. Way inform the readers about the advantages of forecasting and monitoring. Knowing the expected measure of damages caused by insects we can decide whether it is necessary — and if so when to use one or another chemical. For establishing the dangerous number and estimating the loss of yield several methods are described.

The *second chapter* Cultural Methods: The Plant was written by H. F. Van Emden who acquaints the readers with the role of hereditary resistance in plant protection and with the viewpoints whereby the plants can be selected for resistance to pests. The author also describes the generally used methods of transmitting the hereditary resistance.

The *third chapter* "Cultural Methods: The Crop" was written by T. H. Coaker. The author describes some practical control methods useful in the course of cultivation, and calls attention to the importance of site conditions (soil type, climate altitude, etc.). The loss of yield can be substantially reduced when the simple methods generally used before the appearance of the synthetic pesticides are employed. The pests can be prevented from reaching the crop by isolation, further by choosing the optimum time of sowing and

harvesting and the right distance of planting and sequence of crop rotation. Proper soil cultivation, irrigation, fertilization, cleaning of surroundings, weeding, sowing of alluring plants are also components of the IPM.

In the *fourth chapter* S. D. Wratten describes the effectiveness of native natural enemies. The nutrition of predators, such as the *Syrphidae*, *Coccinellidae*, *Chrysopidae*, *Cecidomyidae*, etc. depends on the individual number; and, for the activity of parasites, host organisms are needed in or on which they can lay their eggs.

In the *fifth chapter* I. J. Graham-Bryce deals with "Chemical Methods". The author enumerates the advantages and disadvantages of the protective chemicals: the organochlorine insecticides, organophosphorus insecticides, carbamates, pyrethroids, juvenoids, growth regulators, acaricides, microbial toxins etc., and deals with the methods of application, the forms of packing, the relationship between the physicochemical characteristics of the preparations and their efficiency and with the way of uptake.

"Genetic Control" is the title of the *sixth chapter*, written by E. F. Boller. The autocide method can be easily included in the IPM; excellent results have been achieved with it in the case of the screwworm, *Cochliomyia hominivorax*. Information is given of the sterilization techniques, and of their advantages, disadvantages and basic conditions. The author presents examples of the results attained so far in Central America and Europe.

In the *seventh chapter* E. J. Tait informs the readers of how to organize the IPM. The reduction of pests can be planned according to a system and analysed by computer. Examples are given of how to plan control systems for gemmed shoot, apple pests and late pests of potato.

In the *eighth chapter* A. J. Burn gives an account of the results attained in the integrated protection of cereals. The efficiency of forecasting frit fly, wheat bulb fly, yellow cereal fly, cereal aphids, cereal cyst nematode, European corn borer, gall midge is shown in figures. A survey is provided of the



results of biological and chemical control and of the effects of crop rotation, sowing time, early and late sowing.

In the *ninth chapter* S. Finch describes methods of integrated protection against pests of horticultural crops, such as bean, cabbage, carrot, lettuce, onion and pea, and tells about the results.

P. C. Jepson, the author of the *tenth chapter* "Sugar-Beet" enumerates the major pests and protection techniques. In the case of this crop the main problem is caused by the viruses spread by aphids. The vectors are icontrolled with properly timed aphicides.

The *eleventh chapter* "Fruit and Hops" was written by M. G. Solomon. Informations are given of the major pests of apple, pear, raspberry and hops, which are controlled in practice first of all with chemicals of wide action range. In the case of employing integrated protection based on forecasting, by sparing the predators and parasites the number of sprayings can be reduced and the development of resistance avoided.

In the *twelfth chapter* D. Wainhouse writes about the importance of forestry pests and escribes traditional and modern methods of protection. The use of pheromones, the natural enemies and the microbiological control play a great role in the protection of forests as well.

In the *thirteenth chapter* H. J. Gould sums up the results of control operations against pests of plants grown in glasshouses and under plastic cover. Successes have been achieved with the predatory mite against, two-spotted red spider mite, with parasites against glasshouse whitefly, aphids and leaf-miners, etc. with an entomophagous fungus against aphids and the glasshouse whitefly again, and with the *Bacillus thuringiensis* against caterpillars. Other harmful organisms can be controlled with selective chemicals.

On the ways of reducing quantitative and qualitative losses of stored crops detailed information is given by D. E. Evans in the *fourteenth chapter*. A basic requirement is to store produce that is free from contamination in disinfected areas of suitable temperature and humidity. Pests that appear despite

the above conditions can be destroyed by fumigation, irradiation, with insecticides or with mechanical and biological methods mentioned in the chapter.

The book is completed by an alphabetic "Index" of plants, pests, pathogens, pesticides and plant protection methods which greatly facilitates its use.

The authors did excellent work when using the relevant international literature in this, their book on pest control. No similar comprehensive work has appeared so far. It is particularly important in these days when there is an urgent need for environment protection and cost-saving plant protection techniques. The book will certainly be valuable for those engaged in research, education or crop production; since, when translated into various languages, its conclusions can be employed in all corners of the world.

GIZELLA ÖRDÖGH

*Soil Biology and Conservation of Biosphere.*  
Editor: J. SZEGI, Akadémiai Kiadó, Budapest, 1987.

The 9th International Symposium on Soil Biology and Conservation of Biosphere organized by the Society for Soil Science was held on 27–30 August 1985 at the University of Forestry and Timber Industry, Sopron. The material of the symposium was published by the Publishing House of the Hungarian Academy of Sciences under the editorship of J. Szegi, in English, on 940 pages.

The large number of lectures (104) and the high proportion of participants from abroad show the great interest in these subjects nowadays. In the two volumes of this publication, the subjects are grouped in the following eight chapters:

- Effect of nutrition on the biological processes of the soil
- Interactions of pesticides and soil organisms
- Importance of biological nitrogen fixation in soil fertility



- Role of soil organisms in the decomposition and synthesis of the organic matters of soil
- role of soil organisms in the processes of soil formation
- Soil organisms and their role in the soil ecosystem
- Relationship between soil properties and biological activity
- Interactions of higher plants and soil organisms.

It is worth noting the distribution of the lectures among the chapters. Twenty-three deal with the biological nitrogen fixation, 18 of them with the role of soil organisms in the decomposition and synthesis of the organic matters of soil, 16 discuss the effect of nutrition on the biological processes of the soil. In the remaining 5 chapters, the number of publications ranges between 8 and 12.

In the publication there are lectures based on long term experiments, which offer a broad view encompassing almost the whole branch of knowledge, and give the direction of further investigations; but most of the lectures deal with concrete problems, some smaller part of the subject, which the editor placed in one or another of the above chapters in accordance with their respective subjects.

In the chapter *The effect of fertilization on the soil biological processes* the majority of the 16 lectures discuss the effect of mineral fertilization. According to Zvagintsev and Guzev, relatively low rates of K and N fertilization had a stimulative effect on the microorganisms. However, with an increase in the doses multiplication of the toxin-producing microorganisms must be reckoned with, it is therefore important to determine the maximum rate of fertilization. The resistance of soils to increasing doses of fertilizer greatly depends on the type of soil and the previous method of cultivation. Soils exposed to repeated high rate fertilization showed lower resistance.

Irrigation and high rate fertilization upset the biological balance of the soil, which is manifested in a sudden increase in the num-

ber of certain microorganisms and decrease in that of others — said Dulgerov and Seraya.

The N mineralization of the soil was dealt with by Kovács who pointed out that the values concerning mineralization mostly were underestimated in the literature. Németh and Szebeni discussed the dynamics of N on the basis of the results they obtained in long-term- and culture pot experiments.

In their lecture on enzyme activity Rankov and Dimitrov pointed out that the fertilization had a greater effect on the urease-, protease- and catalase activity than on the activity of asparaginase. Garami and Hargitai found a close correlation between the fertilizer induced amidase activity and the hydrolysable N content.

Bielek and Kudlickova studied the effectiveness of N-Serve in laboratory and lysimeter experiments. Helmecci examined the effect of environment-sparing nutrient bars on the microorganisms.

In the introductory lecture of the chapter *Interactions between pesticides and soil organisms* Takai et al. present some very interesting data on chemicals used in Japan. In long-term field trials they employed insecticide- and pesticide treatments, and studied the effect of adding straw compost. They found that the fungicides had an intensive stimulative effect on the reproduction of bacteria, particularly that of the gramme negative ones. At the same time, their application considerably decreased the number of sporogenous bacteria, fungi and actinomyces, as well as the respiration of soil.

Ananyeva modified the examination technique of the effect of pesticides on microorganisms and determined the microbial biomass effect of pesticide. She obtained reliable changes in treatments with various pesticides.

According to Hickish and Oahn the animals living in the soil are more sensitive to herbicides than the bacteria and fungi.

For the removal of herbicide residues Kunc suggests inoculating the soil with microorganisms able to decompose the herbicides.



Importance of biological nitrogen fixation in soil fertility. Mishustin, in his lecture, pointed out that, an economical increase of yield and soil fertility with the environment spared at the same time cannot be achieved without the knowledge and application of biological nitrogen fixation.

Soybean, as a highly important and valuable source of protein was the subject of a number of lectures. Bonantseva studied the effect of *Spirillum* on the establishment of a symbiosis between soybean varieties and *Rhizobium japonicum*. Kungl examined the acetylene reduction activity in two soils in response to NPK fertilization and inoculation, respectively. Various soybean varieties and *Rhizobium japonicum* strains were studied in Cuba by Piejira; the effect of N- and P fertilization was examined in Egypt by El Shinawi.

Abdalla et al. carried out experiments with lucerne to study the effect of NPK fertilization on the nitrogenase activity. Gulyás and Abdalla employed their own method in examining the nitrogen fixation and found considerable differences between the treatments. Also they studied the seasonal changes of N-fixation. Mareckova found considerable differences in N-fixing capacity for eleven lucerna varieties.

Höflich described the results of examining the effect of *Rhizobium* treatments in various soils in the GDR.

The response of N-fixation to fungicides was examined by Heinonen and Tanski, and to some pesticides by Köves and Péchy et al.

The effects of mineral and organic fertilizer nutrition was dealt with by Brook et al. who emphasized the advantages of organic fertilizer.

#### *Role of soil organisms in the decomposition and synthesis of organic matters*

In Europe the views on the question of organic matter management have changed, and investigations concerning the decomposition and synthesis of organic matters have come into prominence — pointed out Novak in his introductory lecture.

The effect of mineral fertilizer and organic manuring on the organic matter formation and the humification processes was studied by Hargitai in longterm experiments; he found that high rates of mineral fertilization and manuring nutrition, respectively, resulted in humus accumulation.

The role of anaerobic bacteria in humus decomposition was examined by Emtsev and Tuyev; the dynamics of fulvic acid was studied by Murzakov.

The N content of the humus depends on the hydrological conditions under which the process of humification takes place according to Drozd.

Soils respiration and enzyme activity during the decomposition of maize stalks were studied by Tóth. Panteva examined the effect of straw fertilization on the humus components and the biological activity of soil.

The composts and the processes of compostation are also included in this chapter. Mishustin et al. examined the nutritive effect of rice plant remnants. Novakova studied the effect of clay and compost on some characteristics of the organic matter content of the soil. Kanazava and Tsuru followed the biochemical changes important in mushroom cultivation and compostation.

Gulyás et al. studied the effect of communal sewage-sludge on the microbiological processes of soil.

#### *Role of soil organisms in the processes of soil formation*

In this chapter there are lectures by several authors on the effect of microorganisms on soil formation over the spoil-banks of deep- and surface mines. Trofimov et al. determined the successive microbe associations on the basis of 15 years of lysimeter experiments. On the basis of experiments set up on the spoil-banks of the mine at Gyöngyös Visona (North Hungary) Buti et al. described changes in the composition of a *Streptomyces* association. Vörös et al. made interesting statements on the utilization of spoil-banks.

Kuzyurina examined the decomposition



of mineral matters under extreme conditions; Groundeva and Groundev studied the silicate-decomposing bacteria. Włodarczyk discussed the role of *Arthrobacter*- and *Micrococcus* strains.

Buday et al. inoculated an oil-contaminated soil with hydrocarbon-decomposing microbe suspension, and a year later found a great decrease in the oil content.

#### *Soil organisms and their role in the ecosystem*

This chapter includes the material of 8 lectures. The role and importance of the soil microflora is dealt with by Andreyuk et al. An agriculturally cultivated and an intact area with chernozem soil were compared for microorganism activity. The productivity of nitrogen fixation and the intensity of N mobilization were higher on the uncultivated area than on the soil under agrocenosis. The uncultivated area was characterized by humus formation, the cultivated one by humus decomposition.

Semenov established the basic physiological parameters of the prostheobacteria of soil.

Grunda examined the biomass production of fungi by comparing two methods and found that the microscope method gave larger quantities.

Pryczkova studied the effect of fertilization and agronomical practices on the humus- and nitrogen content of the soil under different agrogeological conditions.

Pántos-Derimova dealt with the enzyme activity in the soils of various forest ecosystems.

#### *Relationship between soil properties and biological activity*

This chapter includes the material of 10 lectures. Szegi et al. elaborated a model for characterizing the biological activity of the soil on the basis of the kinetics of respiration and the mathematical analysis of the kinetic curves.

Under the title *Effect of NPK nutrients on the saccharase activity on various Hunga-*

*rian soils*. Anton and Antal pointed out that the positive effect of the treatment was only demonstrable in the presence of cellulose, otherwise no significant change was observed.

In experiments carried out with sewage sludge Anton and Antal found that the dangerous level of heavy metal was better characterized by the saccharase activity than by the total number of microbes.

According to Helmeczi's investigations several years of high rate fertilization caused the pH of soils to fall by nearly a whole degree. Parallel to this the number of bacteria decreased, and the number of microscopic fungi increased. However, in response to liming the number of bacteria grew. On meadow soils a full-, on brown forest soils a half dose of liming was the most efficient.

Velazco et al. examined the effect of liming on 3 soil types. With an increase in the number of bacteria a simultaneous decrease in that of fungi was observed the processes of nitrification became more intensive, and yield increase was observed too.

Oláh-Zsupps and Helmeczi carried out experiments to clear up the effects of soil meliorants.

In the chapter *Interactions of higher plants and soil organisms* Wagner and Buyanovsky deal with the respiration of soils, and discuss the processes resulting in CO<sub>2</sub> formation during the cultivation of winter wheat and the subsequent decomposition of plant parts.

The mycoflora of *Globodera rostochiensis* isolated from soil was the subject of Wronkowska and Janowitz' study.

A serological comparison of *Erwinia Carotovora* isolated from Egyptian and Hungarian soils was carried out by Tohamy and Sárvári.

Mycorrhiza relations were studied by Murontsev et al. with oat and barley; seasonal changes in the growth and activity of mycorrhizal fungi were followed by Ligetfalusi—Kovács.

It is hoped that even this brief review is capable of showing how the relevant values of this publication can benefit the specialists in this field.

MONIKA TAKÁCS





# Weed Research

*Journal of the European Weed Research Society*

**Edited by R.J. Hance** 51 Brook Hill, Woodstock, Oxon OX7 1XH

*Weed Research* is an international journal which publishes papers on all aspects of weeds, their control and related topics. The coverage includes:

- the biology of weeds;
- interactions between weed and crop plants;
- herbicides – their application, formulation, metabolism, mode of action, field performance and environmental fate;
- biological and other control methods;
- agricultural and ecological consequences of weed control practices.

*Weed Research* is the official journal of the European Weed Research Society but authors do not have to be members of the society and papers reporting work done outside Europe, including tropical and subtropical regions, are welcomed. Papers are published in English, French and German with summaries in all three languages.

## Subscription Information

*Weed Research* is published bi-monthly. Subscription rates for 1989 are £72.50 (UK), \$150.00 (USA & Canada), £87.00 (elsewhere) post free.

## Order Form

Please tick the appropriate box and return to Blackwell Scientific Publications Ltd, P.O. Box 88, Oxford, England.

- ☐ I would like to subscribe to *Weed Research*
- ☐ I wish to pay by cheque/money order (*delete as necessary*) and enclose the sum of .....
- ☐ I wish to pay by Access/Barclaycard/VISA/Mastercard (*delete as necessary*)

Please debit my credit card no.

--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--

Expiry date..... with the sum of .....

Signature..... Date .....

- ☐ Please send me a specimen copy of *Weed Research*

Name.....

Address.....

**Blackwell Scientific Publications**

P.O. Box 88, Oxford, England

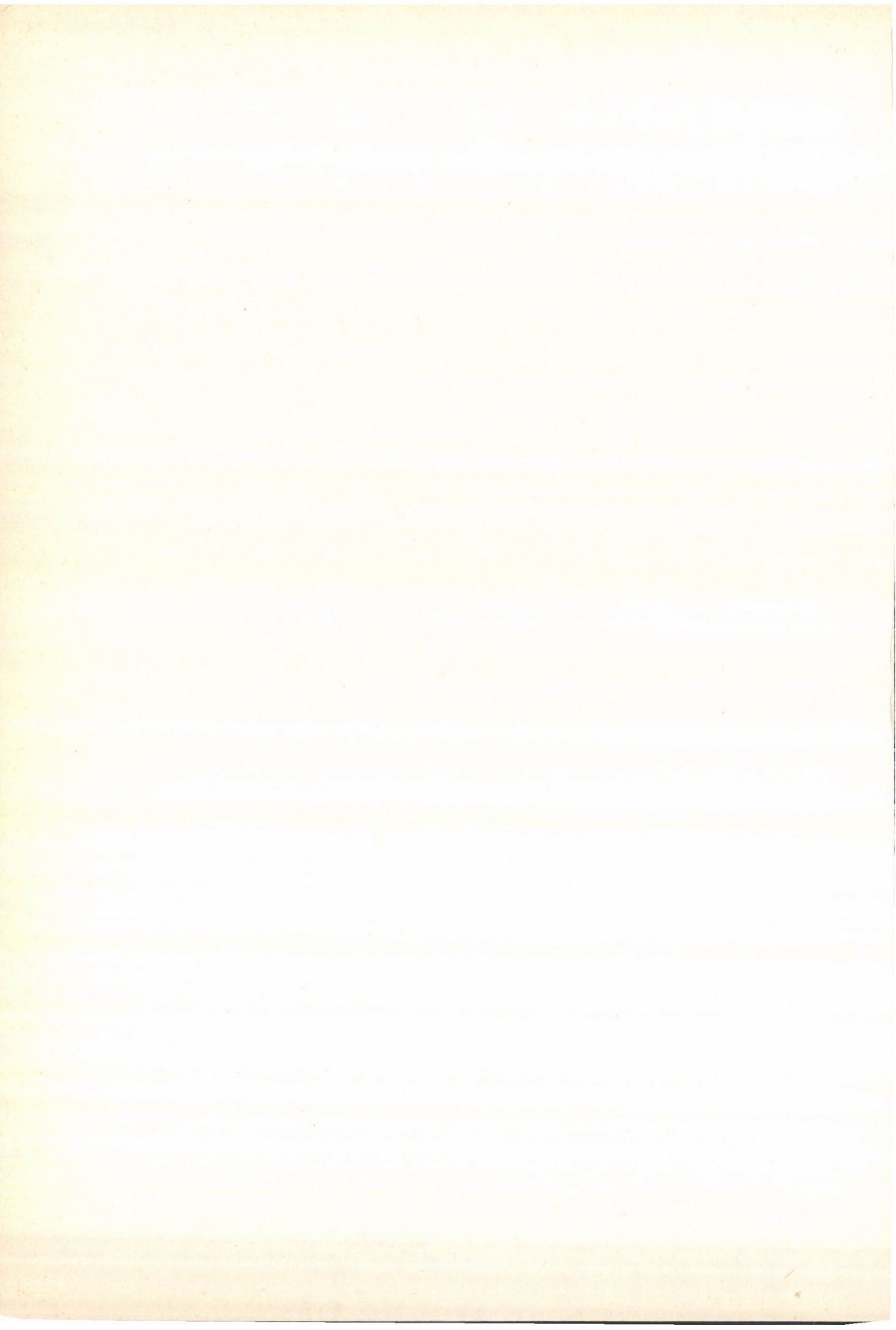




PRINTED IN HUNGARY

Akadémiai Kiadó és Nyomda Vállalat, Budapest





# AUTHORS' GUIDE FOR MANUSCRIPT PREPARATION

## GENERAL INSTRUCTION

Two copies of the manuscript and two sets of the figures should be submitted to:

Acta Agronomica Editorial Office,  
Ménesi út 44.  
H-1118, Budapest

Manuscripts in English or in Hungarian including Abstract, References, Tables and Legends should be typed double-spaced (25 lines, 50 characters per line including spaces) and supplied with authors' names, page number. Tables should be on separate, numbered pages after the References. Legends for figures, on a separate page, should follow the tables. Standard articles should not exceed seven pages.

## FORMAT

*Title.* The title should reflect the most important aspects of the article, in a preferably concise form of not more than 100 characters and spaces.

*By-line.* The authors' names should be followed by affiliations and addresses. (No inclusion of scientific titles is necessary.)

*Abstracts* are required for all the manuscripts. They should be typed in one paragraph and limited to max. 200 words. Below the abstracts, an alphabetical list of keywords should be given.

*Text.* Major sections after the introductory statements are: *Material and methods*, *Results*, *Discussion*, *References*. Subheadings may be used, though the unnecessary fragmentation of the text should be omitted.

*Style.* After acceptance for publication, manuscripts are reviewed for style, grammar and clarity of presentation.

Units should be conform to the International System of Units (SI).

Authors can facilitate editing work by indicating in pencil, the precise meaning of certain symbols (e.g.: distinguish 0 from zero, the number 1 from the letter "l", the multiplication  $\times$  from letter X).

*Names.* Underline Latin binomials to indicate italic type.

*Figures.* Line-drawings should be clear and of high quality. Cite all figures in numerical order in the manuscript. Captions should describe the contents so that each illustration is understandable when considered apart from the text. Each illustration should be labelled with the figure number, author's name, and *Acta Agronomica*.

High-quality glossy prints of photographs should be cropped at right angles to show only essential details. Insert a scale bar where necessary to indicate magnification. Submit two sets of prints of equivalent quality.

*Tables.* The title should be self-explanatory and include enough information so that each table is intelligible without reference to the text or other tables. The title should summarize the information presented in the table without repeating the subheadings. Subheadings should be brief (abbreviations are acceptable) nonstandard ones can be explained in footnotes. Cite tables in numerical order in the manuscript. Information presented in a table should agree with that in the text.

*References.* List literature cited in alphabetic order by authors' surnames. The list should contain names and initials of all authors (et al. is not accepted here); for *journal articles* year of publication, the title of the paper, title of the journal abbreviated (do not abbreviate one word titles), volume number, first and last page. Russian titles should be transliterated and Hungarian titles translated in parentheses.

For books or chapters of books, the titles are followed by the publisher as well as place and date of publication.

Examples:

Kis, Gy., Papp, I., Bakondi-Zámori, É., Gartner-Bánfalvi, Á. (1977): A szőja fungicides magcsávázásának és rhizóbium oltásának együttes tanulmányozása (Joint study of fungicide dressing and rhizobium inoculation in soybean). *Növénytermelés*, **26**, 147-153.

Zinovev, L. S., Matalova, T. S. (1976): Protaviteli, bezopasnie dlya klubenykovykh bakterii. *Zashchita Rastenii*, **5**, 29-31.

Mather, K. and Jinks, J. L. (1971): *Biometrical genetics*. Chapman and Hall Ltd., London, U. K.



Periodicals of the Hungarian Academy of Sciences are obtainable  
at the following addresses:

**AUSTRALIA**

C.B.D. LIBRARY AND SUBSCRIPTION SERVICE  
Box 4886, G.P.O., Sydney N.S.W. 2001  
COSMOS BOOKSHOP, 145 Ackland Street  
St. Kilda (Melbourne), Victoria 3182

**AUSTRIA**

GLOBUS, Höchstädtplatz 3, 1206 Wien XX

**BELGIUM**

OFFICE INTERNATIONAL DES PERIODIQUES  
Avenue Louise, 485, 1050 Bruxelles  
E. STORY-SCIENTIA P.V.B.A.  
P. van Duyseplein 8, 9000 Gent

**BULGARIA**

HEMUS, Bulvar Ruszki 6, Sofia

**CANADA**

PANNONIA BOOKS, P.O. Box 1017  
Postal Station "B", Toronto, Ont. M5T 2T8

**CHINA**

CNPICOR, Periodical Department, P.O. Box 50  
Peking

**CZECHOSLOVAKIA**

MAD'ARSKA KULTURA, Národní třída 22  
115 66 Praha  
PNS DOVOZ TISKU, Vinohradská 46, Praha 2  
PNS DOVOZ TLACE, Bratislava 2

**DENMARK**

EJNAR MUNKSGAARD, 35, Nørre Søgade  
1370 Copenhagen K

**FEDERAL REPUBLIC OF GERMANY**

KUNST UND WISSEN ERICH BIEBER  
Postfach 46, 7000 Stuttgart 1

**FINLAND**

AKATEMINEN KIRJAKAUPPA, P.O. Box 128  
00101 Helsinki 10

**FRANCE**

DAWSON-FRANCE S.A., B.P. 40, 91121 Palaiseau  
OFFICE INTERNATIONALE DE DOCUMENTATION ET  
LIBRAIRIE, 48 rue Gay-Lussac  
75240 Paris, Cedex 05

**GERMAN DEMOCRATIC REPUBLIC**

HAUS DER UNGARISCHEN KULTUR  
Karl Liebknecht-Straße 9, DDR-102 Berlin

**GREAT BRITAIN**

BLACKWELL'S PERIODICALS DIVISION  
Hythe Bridge Street, Oxford OX1 2ET  
BUMPUS, HALDANE AND MAXWELL LTD.  
Cowper Works, Olney, Bucks MK46 4BN  
COLLET'S HOLDINGS LTD., Denington Estate,  
Wellingborough, Northants NN8 2QT  
WM DAWSON AND SONS LTD., Cannon House  
Folkstone, Kent CT19 5EE  
H. K. LEWIS AND CO., 136 Gower Street  
London WC1E 6BS

**GREECE**

KOSTARAKIS BROTHERS INTERNATIONAL  
BOOKSELLERS, 2 Hippokratous Street, Athens-143

**HOLLAND**

FAXON EUROPE, P.O. Box 167  
1000 AD Amsterdam  
MARTINUS NIJHOFF B. V.

Lange Voorhout 9-11, Den Haag  
SWETS SUBSCRIPTION SERVICE  
P.O. Box 830, 2160 SZ Lisse

**INDIA**

ALLIED PUBLISHING PVT. LTD.  
750 Mount Road, Madras 600002  
CENTRAL NEWS AGENCY PVT. LTD.  
Connaught Circus, New Delhi 110001  
INTERNATIONAL BOOK HOUSE PVT. LTD.  
Madame Cama Road, Bombay 400039

**ITALY**

D. E. A., Via Lima 28, 00198 Roma  
INTERSCIENTIA, Via Mazzè 28, 10149 Torino  
LIBRERIA COMMISSIONARIA SANSONI  
Via Lamarmora 45, 50121 Firenze  
SANTO VANASIA, Via M. Macchi 58  
20124 Milano

**JAPAN**

KINOKUNIYA COMPANY LTD.  
Journal Department, P.O. Box 55  
Chitose, Tokyo 156  
MARUZEN COMPANY LTD., Book Department  
P.O. Box 5050 Tokyo International, Tokyo 100-31  
NAUKA LTD., Import Department  
2-30-19 Minami Ikebukuro, Toshima-ku, Tokyo 171

**KOREA**

CHULPANMUL, Phenjan

**NORWAY**

TANUM-TIDSKRIFT-SENTRALEN A.S.  
Karl Johansgata 43, 1000 Oslo

**POLAND**

WĘGIERSKI INSTYTUT KULTURY  
Marzalkowska 80, 00-517 Warszawa  
CKP I W, ul. Towarowa 28, 00-958 Warszawa

**ROUMANIA**

D. E. P., Bucuresti  
ILEXIM, Calea Grivitei 64-66, Bucuresti

**SOVIET UNION**

SOYUZPECHAT — IMPORT, Moscow  
and the post offices in each town  
MEZHDUNARODNAYA KNIGA, Moscow G-200

**SPAIN**

DIAZ DE SANTOS Lagasca 95, Madrid 6

**SWEDEN**

ESSELTE TIDSKRIFTSCENTRALEN  
Box 62, 101 20 Stockholm

**SWITZERLAND**

KARGER LIBRI AG, Petersgraben 31, 4011 Basel

**USA**

EBSCO SUBSCRIPTION SERVICES  
P.O. Box 1943, Birmingham, Alabama 35201  
F. W. FAXON COMPANY, INC.  
15 Southwest Park, Westwood Mass. 02090  
MAJOR SCIENTIFIC SUBSCRIPTIONS  
1851 Diplomat, P.O. Box 819074,  
Dallas, Tx. 75381-9074  
READ-MORE PUBLICATIONS, INC.  
140 Cedar Street, New York, N. Y. 10006

**YUGOSLAVIA**

JUGOSLOVENSKA KNJIGA, Terazije 27, Beograd  
FORUM, Vojvode Mišića 1, 21000 Novi Sad

# **Acta Agronomica Hungarica**

**VOLUME 38, NUMBERS 3-4, 1989**

**EDITOR-IN-CHIEF**

**I. TAMÁSSY**

**EDITOR**

**Á. MÁTHÉ**

**EDITORIAL BOARD**

**S. RAJKI (Vice chairman), I. DIMÉNY, B. GYŐRFFY, A. HORN,  
Z. KIRÁLY, P. KOZMA, E. KURNIK, I. LÁNG, I. MÁTHÉ,  
I. SZABOLCS**



**Akadémiai Kiadó, Budapest**

**ACTA AGRONOMICA HUNG. HU ISSN 0238-0161**



# ACTA AGRONOMICA

## A QUARTERLY OF THE HUNGARIAN ACADEMY OF SCIENCES

---

*Acta Agronomica* publishes papers in English on agronomical subjects, mostly on basic research.

*Acta Agronomica* is published in yearly volumes of four issues by

AKADÉMIAI KIADÓ

Publishing House of the Hungarian Academy of Sciences

H-1054 Budapest, Alkotmány u. 21.

Manuscripts and editorial correspondence should be addressed to

*Acta Agronomica*

H-1118 Budapest, P.O. Box 53

*Subscription information*

Orders should be addressed to

KULTURA Foreign Trading Company

H-1389 Budapest P.O. Box 149

or to its representatives abroad

---

*Acta Agronomica Hungarica* is abstracted/indexed in AGRICOLA, Biological Abstracts, Bibliography of Agriculture, Chemical Abstracts, Current Contents-Agriculture, Biology and Environmental Sciences, Excerpta Medica, Horticultural Abstracts, Hydro-Index, Plant Breeding Abstracts, Nutrition Abstracts and Reviews

---

© Akadémiai Kiadó, Budapest

# ACTA AGRONOMICA HUNGARICA

EDITOR-IN-CHIEF  
I. TAMÁSSY

EDITOR  
Á. MÁTHÉ

EDITORIAL BOARD  
S. RAJKI (Vice chairman), I. DIMÉNY, B. GYÖRFFY, A. HORN, Z. KIRÁLY,  
P. KOZMA, E. KURNIK, I. LÁNG, I. MÁTHÉ, I. SZABOLCS

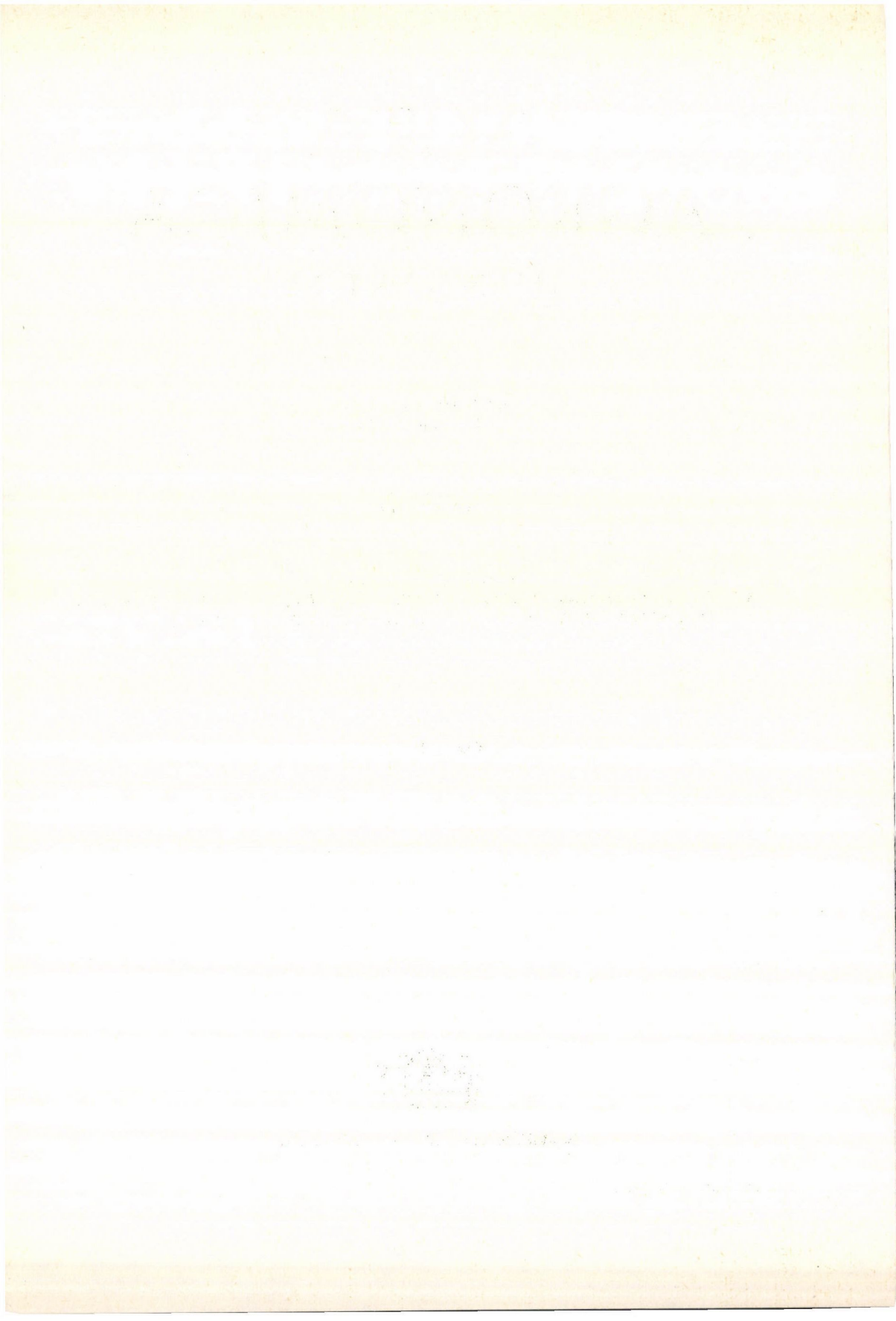
VOLUME 38



AKADÉMIAI KIADÓ, BUDAPEST  
1989

ACTA AGRON. HUNG.





## CONTENTS

NUMBERS 1-2

### SOIL SCIENCE AND AGROCHEMISTRY

Application of electro-ultrafiltration (EUF) method in soil phosphorus determination <i>M. Iqbal Makhdum, M. Nawaz, A. Malik and Fazal Illahi</i> .....	3
Distribution of native fixed ammonium and its prediction as based on textural components and organic carbon in some subtropical soils in West Bengal and Bihar (India) <i>A. Das and B. Datta</i> .....	9

### PLANT PHYSIOLOGY AND BIOCHEMISTRY

Effect of magnesium ion on the anion uptake of plants <i>S. A. Kiss</i> .....	23
Study of flowering in pepper ( <i>Capsicum annum</i> L.) grown under controlled environments (phytotron) <i>A. Máthé and K. L. Bahadli</i> .....	31
Ways of predicting storage losses in apples <i>P. Sass and Z. Lakner</i> .....	37
Investigation of the chemical composition-changes in horticultural plants as a function of X-ray stimulation doses <i>A. S. Szabó and M. A. J. Tejeda</i> .....	45
Effect of ionizing radiation on the respiration intensity of pears during storage <i>Mahfouz Al Bachir and P. Sass</i> .....	49
Tolerance of five oil crops to salinary and temperature stresses during germination <i>F. S. El Nakhlavy and M. A. El Fawal</i> .....	59
Effect of kinetin and salinity on osmotic pressure and carbohydrate contents in two crop plants <i>F. M. Salama and A. A. Awadalla</i> .....	67
Interactive effects of water stress, temperature and $\text{NO}_3^-$ concentration on allocation of soluble nitrogen in germinating <i>Bauhinia</i> seeds <i>H. M. El-Sharkawi and K. A. Farghali</i> .....	77

### PLANT CULTIVATION

Phytomass production of medicinal plants in Finland <i>B. Galambosi</i> .....	89
Effects of plot area and <i>Azolla inocula</i> on growth and nitrogen yield of <i>Azolla pinnata</i> when intercropped with rice <i>D. P. Singh and P. K. Singh</i> .....	99

### PLANT GENETICS AND BREEDING

Heterosis and path coefficient analysis in sesame ( <i>Sesamum indicum</i> L.) <i>H. F. Osman</i> .....	105
Utilization of chlorophyll and leaf mutants in $F_1$ hybrids of watermelons and muskmelons <i>K. Mozsár, H. T. Minch and N. Tamássy</i> .....	113
The influence of repeated back-crosses on the productivity of cytoplasmic male sterile lucerne genotypes <i>B. Nagy</i> .....	119



Inheritance of seed colour in mustard	
<i>D. S. Rawat</i> .....	127
Studies on the agronomic and breeding potentials of some interspecific hybrids in <i>Arachis</i> species	
<i>K. O. Marfo</i> .....	131

## PLANT PROTECTION

Incidence of <i>Tarsonemina</i> species in soil samples of certain tomato varieties	
<i>S. M. Abo-Korah</i> and <i>A. A. Younes</i> .....	139
Tomato transplanting and soil <i>Tarsonemia</i> species	
<i>S. M. Abo-Korah</i> and <i>A. A. Younes</i> .....	143

## ANIMAL PRODUCTION AND GENETICS

Studies on protein utilization of alkaloid-free <i>Luteus albus</i> seed	
<i>J. Jécsai, M. Szelényi-Galántai</i> and <i>B. Juhász</i> .....	149

## LECTURES

Recent results in melon breeding	
<i>K. Mozsár</i> .....	159
Influence of etological factors in establishing the genetic value of mechanical milkability	
<i>L. Szajkó</i> .....	163

## REVIEW

Developments in the trace elements research	
<i>I. Pais</i> .....	167

BOOK REVIEWS .....	177
--------------------	-----

## NUMBERS 3-4

## SOIL SCIENCE AND AGROCHEMISTRY

Elemental content of Hungarian cropland soils using the Mehlich No. 3 extracting reagent and an ICP spectrometer	
<i>J. Benton Jones</i> .....	201

## PLANT PHYSIOLOGY AND BIOCHEMISTRY

Cyclodextrins decrease the phytotoxicity of nonionic tensides	
<i>G. Oros, T. Cserhádi</i> and <i>J. Szejtli</i> .....	211
Antagonism of magnesium and aluminium in bean and wheat	
<i>S. Á. Kiss</i> .....	219
Purification and characterization of myosin from isolated chloroplasts of spinach leaves ( <i>Spinacea oleracea</i> L.)	
<i>R. Nehéz, S. Fazekas, É. Sárvári, I. Óváry</i> and <i>V. Székessy-Hermann</i> .....	231
Effect of fertilization on nutrient uptake and distribution in winter wheat during vegetation	
<i>B. Lásztity</i> .....	241
The rootstock as a modifying factor in the flower organization of the C. 970 Besztercei plum	
<i>D. Surányi</i> .....	255
Response of wheat ( <i>Triticum aestivum</i> L.) treated with cycocel under water stress conditions	
<i>M. Yasin Ashraf, N. A. Baig</i> and <i>F. Baig</i> .....	265
Interactive effects of salinity and phytohormones on growth and plant relationship parameters in maize and safflower plants	
<i>A. F. Radi, M. M. Heikal, A. M. Abdel-Rahman</i> and <i>B. A. El-Deep</i> .....	271

Salinity-hormone interaction in relation to the chemical composition of maize and safflower plants <i>A. F. Radi, M. M. Heikal, A. M. Abdel-Rahman and B. A. A. Deep</i> .....	283
Effect of thinning and halving on grain development in triticale <i>M. R. Rao and V. K. Khanna</i> .....	299
Responses of five forage crops to temperature and salt stresses at germination <i>M. A. El-Fawal and F. S. El Nakhlawy</i> .....	305
<b>PLANT CULTIVATION</b>	
Selection of pollinating plum varieties and their placement in the orchard <i>Z. Szabó and J. Nyéki</i> .....	313
Effect of dolomite- and lime treatment on some quality parameters of Jonathan apples <i>J. Papp and A. H. Aziz</i> .....	331
The effect of different N-doses on changes in the nitrate, sugar and carotene contents of carrot <i>I. Cserni, K. Prohászka and I. Patócs</i> .....	341
The effect of plastic tunnel orientation on yield of some cucumber varieties in Egypt <i>F. El-Aidy</i> .....	349
Effect of Fe and Mn application on the yield and their content in berseem ( <i>Trifolium alexandrinum</i> ) grown in an alkaline soil <i>R. L. Bansal, D. S. Chahal and P. N. Takkar</i> .....	353
<b>PLANT GENETICS AND BREEDING</b>	
Ability of various mutagens to induce chromosomal aberrations in peas <i>Vo Hung</i> .....	357
Inheritance of flag-leaf area in two intervarietal crosses of bread wheat ( <i>Triticum aestivum</i> L.) <i>J. S. Bijral, K. S. Kanwal, B. B. Gupta, B. Singh and T. R. Sharma</i> .....	367
<b>PLANT PROTECTION</b>	
Comparative study of the phytotoxicity of acetanilide herbicides on maize ( <i>Zea mays</i> L.) as affected by temperature and antidotes <i>Z. Berzsenyi and B. Györfy</i> .....	371
On the presence of fusicocein in higher plants <i>G. S. Muromstev, V. D. Voblikova, N. S. Kobrina, V. M. Koreneva, V. L. Sadoskaya and V. V. Stolpakova</i> .....	385
<b>ANIMAL PRODUCTION AND GENETICS</b>	
Composition of colostrum from goats and ewes dropping twins <i>J. Csapó, Gy. Wolf and Zsuzsánna Csapó</i> .....	395
<b>LECTURES</b>	
Soil productivity and some problems of international collaboration and education <i>I. Szabolcs</i> .....	403
<b>REVIEWS</b>	
The importance of plant breeding in the results of crop production with special regard to lucerne <i>I. Bócsa</i> .....	407
Recent problems of amelioration of saline and alkali soils <i>I. Szabolcs</i> .....	419
<b>BOOK REVIEWS</b> .....	431





## CONTENTS

### SOIL SCIENCE AND AGROCHEMISTRY

Elemental content of Hungarian cropland soils using the Mehlich No. 3 extracting reagent and an ICP spectrometer <i>J. Benton Jones</i> .....	201
--	-----

### PLANT PHYSIOLOGY AND BIOCHEMISTRY

Cyclodextrins decrease the phytotoxicity of nonionic tensides <i>G. Oros, T. Cserhádi and J. Szejtli</i> .....	211
Antagonism of magnesium and aluminium in bean and wheat <i>S. Á. Kiss</i> .....	219
Purification and characterization of myosin from isolated chloroplasts of spinach leaves ( <i>Spinacea oleracea</i> L.) <i>R. Nehéz, S. Fazekas, É. Sárvári, I. Óváry and V. Székessy-Hermann</i> .....	231
Effect of fertilization on nutrient uptake and distribution in winter wheat during vegetation <i>B. Lásztity</i> .....	241
The rootstock as a modifying factor in the flower organization of the C. 970 Besztercei plum <i>D. Surányi</i> .....	255
Response of wheat ( <i>Triticum aestivum</i> L.) treated with cycocel under water stress conditions <i>M. Yasin Ashraf, N. A. Baig and F. Baig</i> .....	265
Interactive effects of salinity and phytohormones on growth and plant relationship parameters in maize and safflower plants <i>A. F. Radi, M. M. Heikal, A. M. Abdel-Rahman and B. A. El-Deep</i> .....	271
Salinity-hormone interaction in relation to the chemical composition of maize and safflower plants <i>A. F. Radi, M. M. Heikal, A. M. Abdel-Rahman and B. A. A. Deep</i> .....	283
Effect of thinning and halving on grain development in triticales <i>M. R. Rao and V. K. Khanna</i> .....	299
Responses of five forage crops to temperature and salt stresses at germination <i>M. A. El-Fawal and F. S. El Nakhlawy</i> .....	305

### PLANT CULTIVATION

Selection of pollinating plum varieties and their placement in the orchard <i>Z. Szabó and J. Nyéki</i> .....	313
Effect of dolomite- and lime treatment on some quality parameters of Jonathan apples <i>J. Papp and A. H. Aziz</i> .....	331
The effect of different N-doses on changes in the nitrate, sugar and carotene contents of carrot <i>I. Cserni, K. Prohászka and I. Patócs</i> .....	341



The effect of plastic tunnel orientation on yield of some cucumber varieties in Egypt <i>F. El-Aidy</i> .....	349
Effect of Fe and Mn application on the yield and their content in berseem ( <i>Trifolium alexandrinum</i> ) grown in an alkaline soil <i>R. L. Bansal, D. S. Chahal and P. N. Takkar</i> .....	353

## PLANT GENETICS AND BREEDING

Ability of various mutagens to induce chromosomal aberrations in peas <i>Vo Hung</i> .....	357
Inheritance of flag-leaf area in two intervarietal crosses of bread wheat <i>Triticum aestivum</i> L.) <i>J. S. Bijral, K. S. Kanwal, B. B. Gupta, B. Singh and T. R. Sharma</i> .....	367

## PLANT PROTECTION

Comparative study of the phytotoxicity of acetanilide herbicides on maize ( <i>Zea mays</i> L.) as affected by temperature and antidotes <i>Z. Berzsenyi and B. Györfy</i> .....	371
On the presence of fusicoccin in higher plants <i>G. S. Muromtsev, V. D. Voblikova, N. S. Kobrina, V. M. Koreneva, V. L. Sadovskaya and V. V. Stolpakova</i> .....	385

## ANIMAL PRODUCTION AND GENETICS

Composition of colostrum from goats and ewes dropping twins <i>J. Csapó, Gy. Wolf and Zsuzsánna Csapó</i> .....	395
--	-----

## LECTURES

Soil productivity and some problems of international collaboration and education <i>I. Szabolcs</i> .....	403
--	-----

## REVIEWS

The importance of plant breeding in the results of crop production with special regard to lucerne <i>I. Bócsa</i> .....	407
Recent problems of amelioration of saline and alkali soils <i>I. Szabolcs</i> .....	419

BOOK REVIEWS .....	431
--------------------	-----

## *Soil science and agrochemistry*

---

### ELEMENTAL CONTENT OF HUNGARIAN CROPLAND SOILS USING THE MEHLICH NO. 3 EXTRACTING REAGENT AND AN ICP SPECTROMETER

J. BENTON JONES, JR.

DEPARTMENT OF HORTICULTURE, UNIVERSITY OF GEORGIA, ATHENS, USA

(Received 29 October 1987; accepted 19 January 1988)

Thirty-two Hungarian cropland soils were assayed for their level of extractable Cu, K, Mg, Mn, Na, P and Zn using Mehlich No. 3. extracting reagent and assaying the obtained extract on an inductively-coupled plasma (ICP) emission spectrometer. The level of extractable elements obtained by this procedure compared favorable with values obtained by other testing procedures used by soil testing laboratories in Hungary. Therefore, the Mehlich No. 3. extracting reagent procedure could be used for assessing the soil fertility status of Hungarian soils. The entire extraction-analysis procedure required to assay the 32 soils, by the Mehlich No. 3 method, was completed in an hour.

**Keywords:** Elemental content, Hungarian cropland soils, ICP, Mehlich No. 3. reagent

#### Introduction

Soil testing has been used for over 40 years as a means of assessing the fertility status of cropland soils. The laboratory procedures for conducting these tests has varied considerably depending on soil type and element(s) assayed as well as the factors of past history and experience. In a 1951 survey of soil testing activity in the United States, Nelson et al. (1951) found that there were over 20 different extracting reagents used to assay for extractable P and exchangeable K in soils, although the Morgan extractant (Morgan 1941) was the most commonly used extracting reagent. By 1973, Jones (1973) discovered in his survey that the number of different extracting reagents had narrowed to essentially four, Bray P1 (Bray and Kurtz 1945) and sodium bicarbonate (Olsen et al. 1954) for extractable P, neutral normal ammonium acetate (Schollenberger and Simon 1945) for determining the exchangeable cations K, Ca and Mg, and Mehlich No. 1 (Mehlich 1953) for the determination of P, K, Ca and Mg contents in the acid sandy soils found in the southeastern United States.

Today, the shift is from single element extracting reagents, like Bray P1, to reagents that can be used for both the major elements as well as the micro-



nutrients. Such extraction procedures can take advantage of multielement analyzers, such as the inductively-coupled plasma (ICP) spectrometer (Jones 1977; Soltanpour, Workman and Schwab 1979; Munter and Grande 1981; Soltanpour, Jones and Workman 1982). The two extracting reagents that are coming into wide use are ammonium bicarbonate-DTPA, frequently referred to as AB-DTPA (Soltanpour and Schwab, 1977) for the determination of P, K, Na, Cu, Fe, Mn and Zn in alkaline soils, and Mehlich No. 2 (Mehlich 1984) for the determination of P, K, Ca, Mg, Na, Cu, Fe, Mn and Zn in acid soils. The elemental content of the extract obtained from the interaction of the extracting reagent with soil is easily and quickly assayed on an ICP spectrometer. Both extractants have been calibrated in terms of defining the nutrient element status of soils as well as providing the basis for making fertilizer recommendations (Hanlon and Johnson 1984; Johnson and Tucker 1985; Soltanpour and Follett 1985).

In addition, both of these extracting procedures employ volume measurement (scooping in lieu of weighing) of the aliquot of soil to be extracted which considerably speeds the testing process. The rationale for volume aliquot measurement has been discussed by Mehlich (1972, 1973). Scoops, their design (diameter being twice their depth) and how they are used are important factors that determine their ability to consistently obtain the desired soil aliquot (Peck 1980). Scooping to obtain either a specific volume of soil or an estimated weight of soil is the common practice in most soil testing laboratories in the United States.

The objective of this study was to compare extractable element content obtained by the Mehlich No. 3. extracting procedure versus those obtained by other assaying procedures in use in Hungarian Soil Testing Laboratories.

### Materials and methods

Thirty-two Hungarian cropland soils were air dried, sieved through a 10-mesh sieve and assayed in the Pelence and Tenakagh laboratories. The soils were assayed for their pH in KCl, percentage of free calcium carbonate, and organic matter contents, level of 0.1 N ammonium lactate in 0.1 N acetate extractable P and K, 1 N potassium chloride extractable Mg and Na, and 0.5 M EDTA in 0.1 N potassium chloride extractable Cu, Mn and Zn. Soil pH ranged from 4.0 to 7.3 with 7 soils having measurable free calcium carbonate contents. Organic matter contents ranged from a low of 2.0% to a high of 4.0%.

The 32 soils were taken to the Department of Horticulture laboratories at the University of Georgia for assaying with the Mehlich No. 3 procedure. Using a volume scoop, an 8.5 cm<sup>3</sup> aliquot of soil was placed into a 250 ml polyethylene Erlenmeyer flask. An 85 ml aliquot of Mehlich No. 3. extracting agent (0.02 N acetic acid in 0.25 N ammonium nitrate in 0.015 N ammonium fluoride in 0.013 nitric acid in 0.01 M EDTA) was added to the 150 ml Erlenmeyer flask and the mixture of soil and extracting agent shaken vigorously for 5 minutes. The soil extractant mixture was filtered using Whatman No. 1 filter paper and the filtrate collected in a polyethylene bottle. The Ca, Cu, K, Mg, Mn, Na, P and Zn contents of the filtrate were determined using a Jarrell Ash Model 750 ICP spectrometer (Jones 1977). There was no need to modify or dilute the filtrate which was assayed as prepared on the ICP spectrometer, since the readable range for the spectrometer is over 4 orders of magnitude. Being a



polychromator ICP, the analysis time was less than 40 seconds per sample for all 8 element determinations. The entire analysis procedure from extraction to final analysis results for all 32 samples took about an hour to complete. Regression figures of elemental concentration obtained by the Mehlich No 3 extraction procedure versus elemental concentration reported by the Hungarian laboratories were drawn for each element using an IBM PC computer program and printer.

### Results and discussion

Calcium results were not given by the Hungarian laboratories but were obtained by the Mehlich No. 3-ICP procedure. In addition, the cation exchange capacity of the 32 soils was calculated based on the level of Mehlich No. 3.

**Table 1**

*Calcium content and calculated cation exchange capacity for 32 Hungarian soils based on the Mehlich No 3 extraction procedure*

Sample number	Calcium content, $\mu\text{g/kg}$	Cation exchange capacity meq/100g
43	3329	23.5
44	3203	23.0
45	3313	23.5
46	3597	25.5
47	3126	22.7
48	5960	37.1
49	5500	37.8
50	7679	44.2
51	4038	29.1
52	3613	29.4
55	7636	41.3
56	6684	36.5
57	3453	23.1
59	3228	25.3
61	6249	38.9
62	1324	17.3
63	833	15.4
64	2176	14.9
65	2193	15.5
67	2352	17.2
68	1665	13.8
69	2779	24.7
70	2856	25.1
71	2988	25.3
72	3667	25.5
73	3275	24.2
74	5105	27.6
78	3378	24.7
79	3136	23.4
94	3274	23.2
95	3107	23.2
96	2945	21.8
99	4243	24.3
100	4097	23.2



extractable bases (Ca, K, Mg and Na) and milliequivalents of hydrogen determined by the Adams-Evans buffer procedure (Adams and Evans 1962). Both sets of values are given in Table 1.

Actual Mn concentrations were reported by the Hungarian laboratories for only 7 soils and therefore no useful comparison could be made between the two sets of assay data. No Mg information was given for 7 soils by the Hungarian laboratories. In both instances, the Mn and Mg concentrations evidently exceeded the readable range for the analytical methods used in the Hungarian laboratories. Actual elemental concentrations of both elements and in all 32 soils assayed were obtained by the Mehlich No. 3 procedure.

A regression between microgram per kilogram ( $\mu\text{g/kg}$ ) concentration for the elements Cu, K, Mg, P, Na and Zn in soil reported by the Hungarian soil testing laboratories, using their extraction and assay procedures, and that obtained by the Mehlich No. 3-ICP procedure was determined. The regression curves and equations are shown in Figures 1 through 7.

The regression between 0.05 M EDTA in 0.1 N potassium chloride versus Mehlich No. 3. extractable Cu was statistically significant ( $r = 0.89^{***}$ ), although the scatter around the regression line was considerable. Copper is a difficult element to extract from soils and its assay can be equally difficult. Use of an outlier program could be applied to improve the regression; however, additional soil assay data would be needed to justify the use of such a procedure. There were four soils in which Mehlich No. 3 gave considerably higher values than that obtained by the Hungarian extracting reagent, which may be due to an analytical error being made in one of the laboratories. A marked

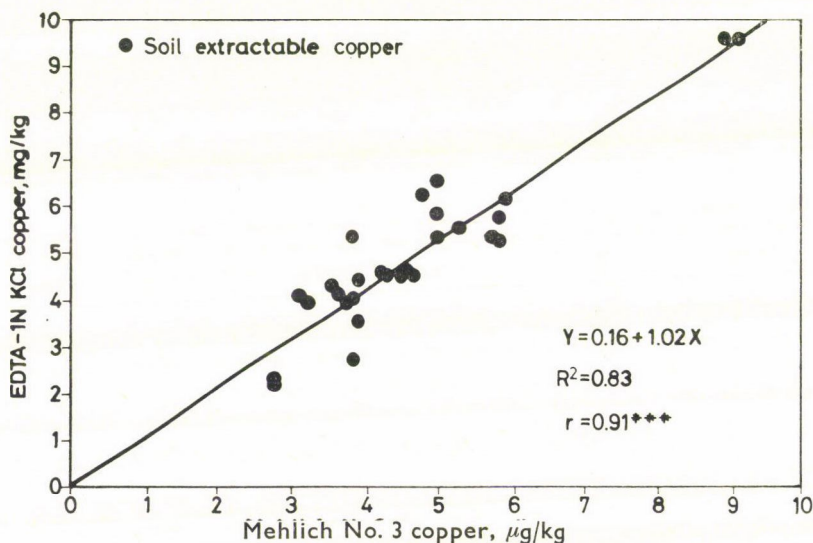


Fig. 1. Regression between 0.05 M EDTA in 0.1 N KCl and Mehlich No. 3 extractable copper

improvement in the significance of the regression ( $r = 0.91^{***}$ ) is obtained if these four soil results are removed from the regression as shown in Figure 1.

The regression between 0.1 N ammonium lactate in 0.1 N acetic acid extractable K versus that extracted by Mehlich No. 3 was highly significant ( $r = 0.97^{***}$ ) with little scatter of data points around the regression line

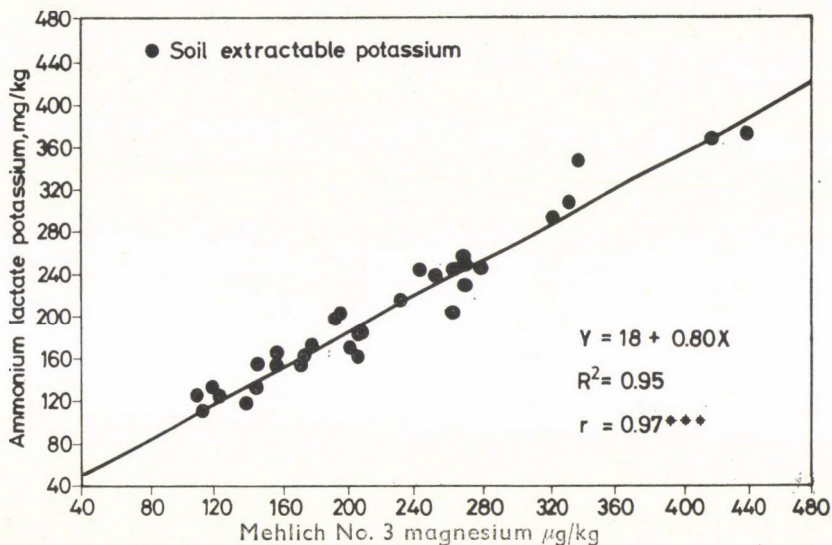


Fig. 2. Regression between 0.1 N ammonium lactate in 0.1 N acetic acid and Mehlich No. 3 extractable potassium

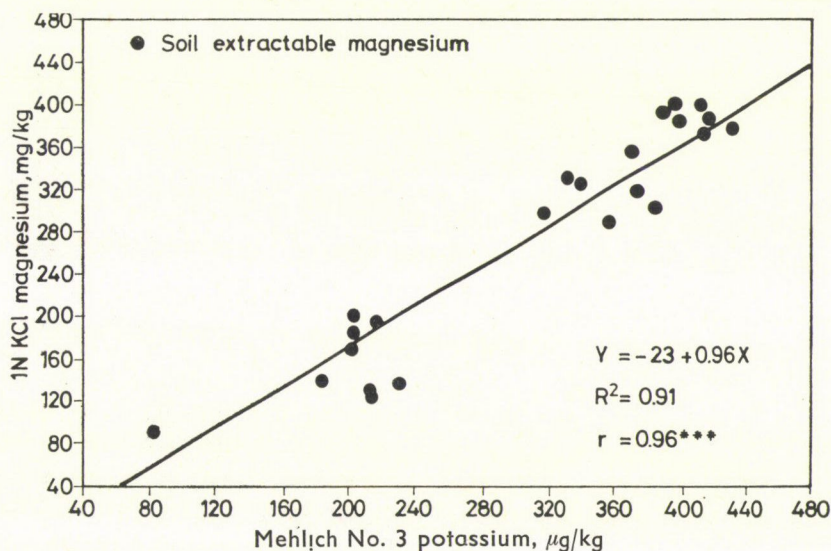


Fig. 3. Regression between 1 N potassium chloride and Mehlich No. 3 extractable magnesium



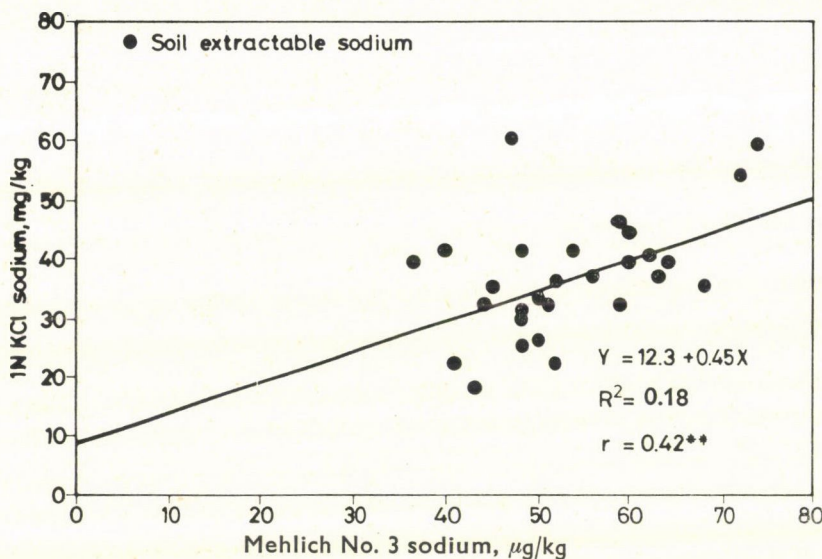


Fig. 4. Regression between 1 N potassium chloride and Mehlich No. 3 extractable sodium

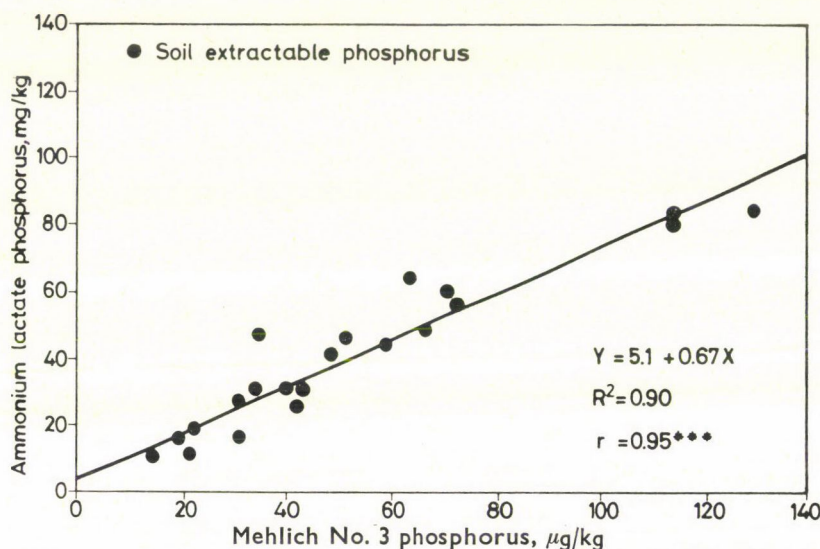


Fig. 5. Regression of 0.1 N ammonium lactate in 0.1 N acetic acid and Mehlich No. 3 extractable phosphorus in acid soils

(Figure 2). The difference in the amount of K extracted by Mehlich No. 3 compared to the lactate extracting reagent increases as the soil K level increases. However, within the sufficiency range (0 to 200  $\mu\text{g K/kg}$ ) for K, both extractants yield essentially the same K concentration.

The regression between 1 N potassium chloride extractable Mg versus that extracted by Mehlich No. 3 was highly significant ( $r = 0.95^{***}$ ) with

some scatter of the data points around the regression line (Figure 3). There seems to be two sets of soils, one lower in Mg content than the other. Additional lower Mg content soils need to be tests compared in order to more closely examine the regression at Mg soil levels below 200  $\mu\text{g Mg/kg}$ .

The regression between 1 N potassium chloride extractable Na versus that extracted by Mehlich No. 3. was statistically significant ( $r = 0.42^{**}$ ), although the scatter around the regression line is considerable (Figure 4). The level of Na in these soils is quite low (less than 100  $\mu\text{g Na/kg}$ ), and therefore, this comparison of methodology is probably not being fairly tested. Errors due to contamination and assay technique are considerable at such low concentration levels.

Even though 7 soils had detectable amounts of free calcium carbonate, the regression of 0.1 N ammonium lactate in 0.1 N acetic acid versus Mehlich No. 3. extractable P was statistically significant ( $r = 0.83^{***}$ ), although there was considerable scatter around the regression line (Figure not shown). Removing these 7 soils from the determination markedly improved the correlation ( $r = 0.95^{***}$ ) and the regression as shown in Figure 5. Surprisingly, the regression for the 7 alkaline soils alone gave a high correlation ( $r = 0.93^{***}$ ) for P extracted by the two methods (Figure 6). It is interesting to note that Mehlich No. 3 determined P concentrations were less than that extracted by ammonium lactate for the generally acid soils, while the amount extracted by the two methods was reversed for the 7 soils with measurable calcium carbonate levels.

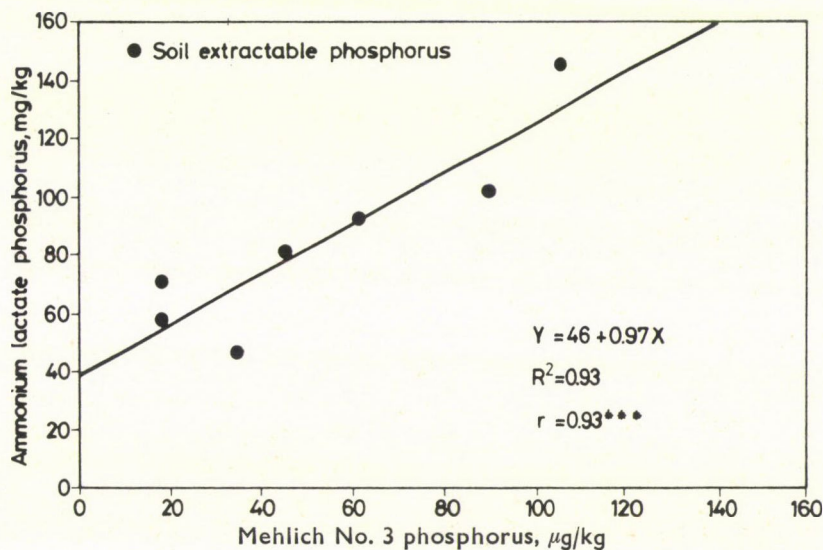


Fig. 6. Regression between 0.1 N ammonium lactate in 0.1 N acetic acid and Mehlich No. 3 extractable phosphorus in 7 soils containing free calcium carbonate



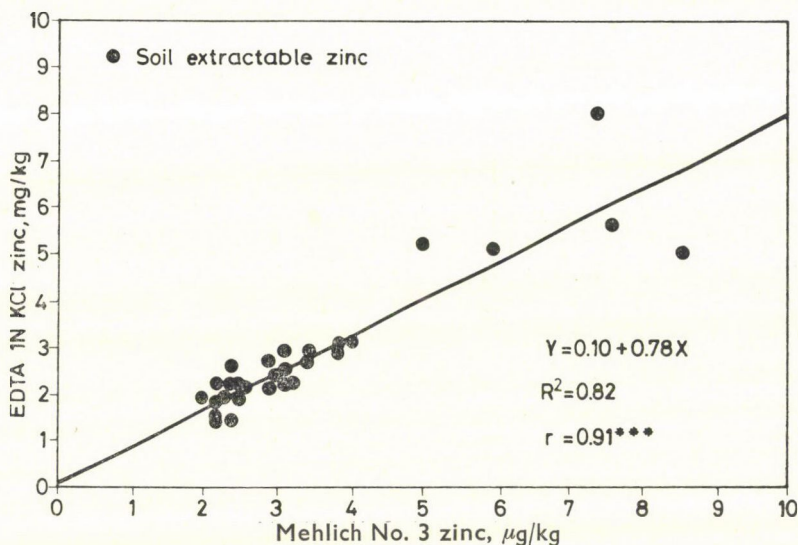


Fig. 7. Regression between 0.05 M EDTA in 0.1 N KCl and Mehlich No. 3 extractable zinc

The regression between 0.05 M EDTA in 0.1 N KCl extractable Zn versus that extracted by Mehlich No. 3 was highly significant ( $r = 0.91^{***}$ ) with little scatter around the regression line at concentrations less than  $5 \mu\text{g Zn/kg}$ . Above this concentration, there were 5 soils whose only common characteristic was their very high P content (greater than  $150 \mu\text{g/kg}$  ammonium lactate extractable). Within the sufficiency range for Zn (less than  $5 \mu\text{g/kg}$ ), both extractions gave essentially the same Zn concentration.

In summary, the Mehlich No. 3 extraction-ICP method gave similar results for the elements Cu and Zn as those obtained by the methods used in the Hungarian laboratories. For the elements K, Mg, Na and P, the concentrations, obtained by the two methods of analysis were significantly correlated. Therefore, the Mehlich No. 3 results could be used in place of the Hungarian laboratory results by either regression transformation, or through calibration of the Mehlich No. 3 elemental concentration values in terms of those used to interpret the Hungarian tests results. Soils would have to be separated on the basis of free calcium carbonate content in order to make the P transformations. As was suggested, additional soils with varying concentrations of Mg and Na need to be compared to ensure the reliability of the association of results between the two assaying methods.

The Mehlich No. 3-ICP analysis procedure was fast and provided elemental concentration data for all elements compared in this study, plus Ca, without the need for extract manipulation.

## References

- Adams, F., Evans, C. E. (1982): A rapid method for measuring lime requirement of red-yellow podzolic soils. *Soil Sci. Soc. Amer. Proc.*, **26**, 355-357.
- Bray, R. H., Kurtz, L. T. (1945): Determination of total, organic and available forms of phosphorus in soils. *Soil Sci.*, **59**, 39-45.
- Hanlon, E. A., Johnson, G. V. (1984): Bray/Kurtz, Mehlich III, AB/DTPA, ammonium acetate extractions of P, K and Mg in four Oklahoma soils. *Comm. Soil Sci. Plant Anal.*, **15**, 277-294.
- Johnson, G. V., Tucker, B. (1985) OSU soil test calibration. Oklahoma State Univ. Ext. Facts No. 2225.
- Jones, Jr., J. B. (1973): Soil testing in the United States. *Comm. Soil Sci. Plant Anal.*, **4**, 307-322.
- Jones, Jr., J. B. (1977): Elemental analysis of soil extracts and plant tissue ash by plasma emission spectroscopy. *Comm. Soil Sci. Plant Anal.*, **8**, 349-365.
- Mehlich, A. (1953): *Determination of P, Ca, Mg, K, N and NH<sub>4</sub>*. North Carolina Division Mimeo.
- Mehlich, A. (1972): Uniformity of expressing soil test results: A case for calculating results on a volume basis. *Comm. Soil Sci. Plant Anal.*, **3**, 417-424.
- Mehlich, A. (1973): Uniformity of soil tests results as influenced by volume weight. *Comm. Soil Sci. Plant Anal.*, **4**, 475-486.
- Mehlich, A. (1984): Mehlich No. 3 soil test extractant: A modification of Mehlich No. 2 extractant. *Comm. Soil Sci. Plant Anal.*, **15**, 1409-1416.
- Morgan, M. F. (1941): Chemical diagnosis by the universal soil testing system. *Conn. Agr. Exp. Sta. Bull.* 450.
- Munter, R. C., Grande, R. A. (1981): *Plant tissue and soil extract analysis by ICP-atomic emission spectrometry*. In: *Developments in atomic emission spectrometry* (ed.) Barnes, R. M. Heyden & Son, LTD, London, 653-672.
- Nelson, W. L., Fitts, J. W., Kardos, L. T., McGeorge, W. T., Parks, R. O., Reed, Fielding, J. (1951): *Soil testing in the United States*. National Soil & Fertilizer Research Committee. Publication No. 0-979953. U. S. Government Printing Office.
- Olsen, S. R., Cole, C. V., Watanabe, F. S., Dean, L. A. (1954): *Estimation of available phosphorus in soils by extraction with sodium bicarbonate*. USDA Circular 939.
- Peck, T. R. (1980): Standard soil scoop. In: *Recommended chemical soil test procedures for the north central region* (ed) Dahnke, W. C. North Dakota Agr. Expt. Sta. Bull. **499**, (rev.), 3-4.
- Shollenberger, C. J., Simon, R. H. (1945): Determination of exchange capacity and exchangeable bases in soil-ammonium acetate. *Soil Sci.*, **59**, 1-24.
- Soltanpour, P. N., Schwab, A. P. (1977): A new soil test for simultaneous extraction of macro- and micro-nutrients in alkaline soils. *Comm. Soil Sci. Plant Anal.*, **8**, 195-207.
- Soltanpour, P. N., Workman, S. M., Schwab, A. P. (1979): Use of inductively-coupled plasma spectrometry for the simultaneous determination of macro- and micro-nutrients in  $\text{NH}_4\text{HNO}_3$ -DTPA extracts of soils. *J. Soil Sci. Amer.*, **43**, 73-78.
- Soltanpour, P. N., Jones, Jr., J. B., Workman, S. M. (1982): Optical emission spectrometry. In: *Methods of soil analysis, Part 2, Chemical and microbiological properties* (2nd ed.) (ed) Page, A. L. American Society of Agronomy, Madison, W. I., 26-65.
- Soltanpour, P. N., Follett, R. H. (1985): Soil test explanation. Colorado Ext. Serv. Action No. 502.





## *Plant physiology and biochemistry*

### CYCLODEXTRINS DECREASE THE PHYTOTOXICITY OF NONIONIC TENSIDES

G. OROS,<sup>1</sup> T. CSERHÁTI<sup>1</sup> and J. SZEJTLI<sup>2</sup>

<sup>1</sup> PLANT PROTECTION INSTITUTE, HUNGARIAN ACADEMY OF SCIENCES,  
BUDAPEST, HUNGARY

<sup>2</sup> BIOCHEMICAL RESEARCH LABORATORY OF CHINOLIN PHARMACEUTICAL  
AND CHEMICAL WORKS, BUDAPEST, HUNGARY

(Received 30 June 1987; accepted 18 November 1987)

Nonionic tensides belonging to the polyethoxylated nonylphenole, tributylphenole, oleylalcohol, and sorbitane derivatives cause phytotoxic symptoms in plants. This action depends on the length of ethyleneoxid chain and on the type of the hydrophobic part of the tensides. Alpha-, beta-, gamma-cyclodextrines, 2,6-di-*O*-methyl- $\beta$ -cyclodextrin and 2,3-tri-*O*-methyl- $\beta$ -cyclodextrin prevent or decrease the phytotoxicity of tensides. This phenomenon may be due to the inclusion and complex formation of tensides with cyclodextrins, resulting in a decreased concentration of free tensides and consequently in a lesser phytotoxicity.

**Keywords:** Age response, collapse of leaf tissue, cyclodextrin, inclusion complex formation, nonionic tensides, phytotoxicity, sugar-beet tobacco

#### Introduction

Pesticide formulations usually contain nonionic tensides that improve such technical parameters as drop volume, suspension or emulsion stability, better spreading on the leaf surface, etc. (Van Valkenburg, 1973). In addition to the above effects, the tensides can modify the biological efficiency of the active substances (Freed and Witt, 1969; Dunleavy et al., 1982; Spotts and Peters, 1982) and even their selectivity (Müller and Burth, 1983). Sometimes they show marked microbicidal (Baicu and Jilaveanu, 1977; Clifford and Hislop, 1975) and phytotoxic (Babiker and Duncan, 1975; Parochetti, Wilson and Beste, 1977; Leroux and Harvey, 1981) effects.

Nonionic tensides have been shown to interact with membrane phospholipids (Cserhádi, Szőgyi and Győrfi, 1986) and increase the membrane permeability (Szőgyi, Tölgyesi and Cserhádi, 1983).

Due to their capacity to form inclusion complexes with a large number of organic compounds, cyclodextrins receive a growing acceptance and application in both human therapy and phytopharmacology (Szejtli, 1982, 1984). Tenside form complexes with cyclodextrins, resulting in the change of the



critical micelle concentration and the surface tension in water solutions (Koch, 1982; Králová et al., 1982; 1983; Okubo et al., 1976). Therefore it is of interest to study the influence of inclusion complex formation on phytotoxic effect of tensides.

### Materials and methods

The chemical structure of commercial grade nonionic tensides are shown in Figure 1. The cyclodextrins:  $\alpha$ -cyclodextrin ( $\alpha$ CD),  $\beta$ -cyclodextrin ( $\beta$ CD),  $\gamma$ -cyclodextrin ( $\gamma$ CD), 2,6-di-*O*-methyl- $\beta$ -cyclodextrin (DIMEB) and 2,3,6-tri-*O*-methyl- $\beta$ -cyclodextrin (TRIMEB)

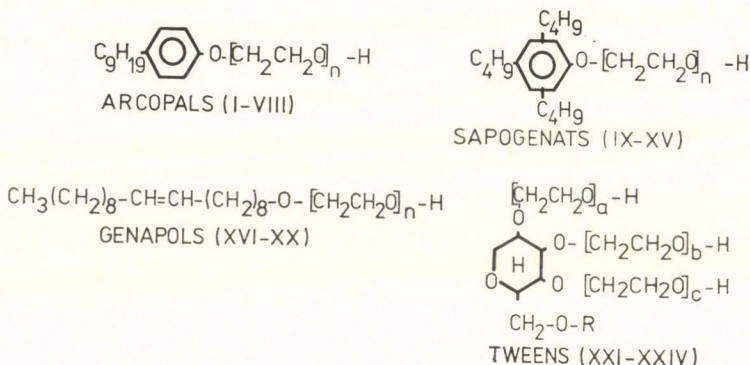


Fig. 1. The structure of tensides. The number of ethoxy groups ( $n$ ) is shown for arcopals (*p*-nonylphenyl glycolates), sapogenates (tributylphenyl glycolates) and genapols (oleyl glycolates), i.e. for compounds I–XX, respectively in Table 1. The number of ethoxy groups ( $a$ ,  $b$ ,  $c$ ) is 20 as a sum for TWEENS (sorbitane glycolate esters), where  $R$  is laurate (TWEEN 20), palmitate (TWEEN 40), stearate (TWEEN 60) and oleate (TWEEN 80) in the compounds XXI–XXIV, respectively

were produced by CHINOIN (Budapest, Hungary) and applied without additional purification.

The plants tested (*Nicotiana tabacum* L. cv. Xanthi, *Beta vulgaris* L. cv. Monopoly N-1, and *Tradescantia albiflora* L.) were grown in a greenhouse in soil culture.

#### Test for phytotoxicity

Tensides dissolved in distilled water at the concentrations of 10, 5, 1, 0.5 and 0.25 mM were infiltrated into leaf sheets of plants (about  $15 \times 10$  mm areas with each tenside at appropriate concentration, respectively). The phytotoxic effect was checked after 12, 24, 48 and 72 hours by the 0–2 scale: 0—no symptoms; 1—the necrosis was not induced, and the infiltrated area became yellow after 48–72 h; 2 — the necrosis was induced in an infiltrated area during 72 h.

For the estimation of protective effect of cyclodextrins, their mixtures with tensides in molar ratios 1 : 1, 1 : 2, 1 : 6 and 1 : 10 (10 mM tenside : cyclodextrin) were tested as above.

Each experiment was carried out in quadruplicate.

#### Data analysis

Both potential toxicity of tensides and age responses of leaf-sheets were calculated from the original data using the Potency Mapping technique, described by Lewi (1976). The relation between potential phytotoxicity (PT) of tensides and the degree of their ethoxylation was analysed by curve fitting. Statistical interpretation was made by reference to Sváb (1981).

Table 1

*Age response of tobacco leaves to varying concentrations of nonionic tensides*  
(Critical concentrations (mM) for induction of leaf collapse)

Tensides*		Leaf position							Potential toxicity %
No	number of ethoxy groups	1	2	3	4	5	6	7	
Arcopalís									
I	4	>10	>10	>10	>10	>10	>10	>10	13.4
II	6	1-5	1-5	1-5	1-5	1-5	1-5	1-5	83.7
III	9	0.5	1-5	1-5	1-5	1-5	1-5	1-5	87.0
IV	10	1	1-5	1-5	1-5	1-5	1-5	1-5	86.2
V	13	0.5	0.5-1	1-5	1-5	1-5	1-5	1-5	83.7
VI	15	0.5	0.5-1	1-5	1-5	1-5	1-5	1-5	83.7
VII	23	5	>10	>10	>10	>10	>10	>10	24.1
VIII	30	>10	>10	>10	>10	>10	>10	>10	13.4
Sapogenats									
IX	4	5-10	5-10	5-10	5-10	5-10	>10	>10	34.1
X	6	0.5-1	1-5	1-5	1-5	1-5	1-5	1-5	86.6
XI	8	0.5-1	1-5	1-5	1-5	1-5	1-5	1-5	86.6
XII	10	1-5	1-5	1-5	1-5	1-5	1-5	1-5	83.4
XIII	13	0.25-0.5	1-5	1-5	1-5	1-5	1-5	1-5	87.5
XIV	18	0.25-0.5	0.5-1	0.5-1	0.5-1	0.5-1	0.5-1	1-5	100
XV	30	0.5-1	1-5	1-5	1-5	1-5	1-5	1-5	86.6
Genapolis									
XVI	2	>10	>10	>10	>10	>10	>10	>10	13.4
XVII	5	>10	>10	>10	>10	>10	>10	>10	13.4
XVIII	8	1-5	5	5-10	5-10	10	>10	>10	43.4
XIX	15	1-5	1-5	1-5	1-5	1-5	1-5	1-5	83.4
XX	20	1-5	1-5	1-5	1-5	1-5	1-5	1-5	83.4
Tweens									
XXI	20	1-5	1-5	1-5	1-5	1-5	5	5	77.7
XXII	20	1-5	1-5	1-5	1-5	1-5	5	5	77.7
XXIII	20	1-5	1-5	1-5	1-5	1-5	1-5	5-10	77.0
XXIV	20	1-5	1-5	1-5	1-5	1-5	1-5	1-5	83.4
Potential sensitivity **(%o)									
		94	88	76	71	47	40	35	

\* = The structure is shown in Fig. 1.

\*\* = Leaf response to the compounds studied is characterized by the weighed averages of potential sensitivities to given groups of tensides.

The significance of differences between age responses were tested by *F* probe (SVÁB, 1981):  $F_{P-10\%} = 1.38 < F_{1,2} = 1.62$   $F_{P-5\%} = 1.65 < F_{1,3-5} = 1.85 < < F_{P-1\%} = 2.04 < F_{1,6} = 2.08 < F_{1,7} = 2.53$ ;  $F_{2,3-6} = 1.13-1.28 < F_{P-10\%} = 1.38 < F_{2,7} = 1.55$ ; the differences between responses of leaves on 3<sup>rd</sup>-7<sup>th</sup> levels over epicotyl are insignificant.



Table 2

*Alteration of plant responses to tensides with cyclodextrins  
(3<sup>rd</sup> and 4<sup>th</sup> level of leaves on tobacco)*

Tenside <sup>a</sup> (10 mM)	Cyclodextrine <sup>b</sup>	Phytotoxic effect			
		Tenside/Cyclodextrin molar ratio <sup>c</sup>			
		1:1	1:2	1:6	1:10
IV*	$\alpha$ -CD	2**	2	2	2
	$\beta$ -CD	2	2	1	0
	$\gamma$ -CD	2	1	0	0
	DIMEB	0	0	0	0
	TRIMEB	0	0	0	0
VI	$\alpha$ -CD	2	2	2	2
	$\beta$ -CD	2	2	2	2
	$\gamma$ -CD	2	2	0	0
	DIMEB	0	0	0	0
	TRIMEB	0	0	0	0
XI	$\alpha$ -CD	2	2	2	2
	$\beta$ -CD	2	2	2	2
	$\gamma$ -CD	2	0	0	0
	DIMEB	2	2	2	2
	TRIMEB	2	2	2	2
XX	$\alpha$ -CD	2	2	0	0
	$\beta$ -CD	2	2	2	2
	$\gamma$ -CD	2	2	0	0
	DIMEB	2	2	2	2
	TRIMEB	2	2	2	2
XXII	$\alpha$ -CD	0	0	0	0
	$\beta$ -CD	0	0	0	0
	$\gamma$ -CD	1	0	0	0
	DIMEB	2	2	2	2
	TRIMEB	2	2	2	2

\* = The structure of tensides is shown in Fig 1.

\*\* = Numbers indicate the degree of damage caused during 24 hours.

The differences between efficacy of various treatments were tested by *F* probe (Sváb, 1981):  $F^a = 15.97 > F^b = 7.68 > F^c = 6.91 > F_{P=0.1\%} = 6.17$ ;  $F^{a,b} = 12.45 > F_{P=0.1\%} = 3.31$ ;  $F^{a,b,c} = 3.73 > F_{P=0.1\%} = 2.23$ .

## Results

The age response of tobacco leaves to tensides is shown in Table 1. The tolerance decreased with the age of leaves, regardless of the tenside type. The response of leaves on the 3<sup>rd</sup> and 4<sup>th</sup> levels over epicotyl to tensides did not differ significantly. The old leaves (1<sup>st</sup> level over epicotyl exhibited about 5 times more sensitivity to the tensides than did the youngest one ( $P = 0.1 - 1\%$ ).

The tensides with low and high number of ethylene oxide groups (*I*, *IX*, *XIV*, *XVII* and *VIII*, respectively) did not exhibit phytotoxicity of practical importance. The most active compounds belong to nonyl-phenyl and

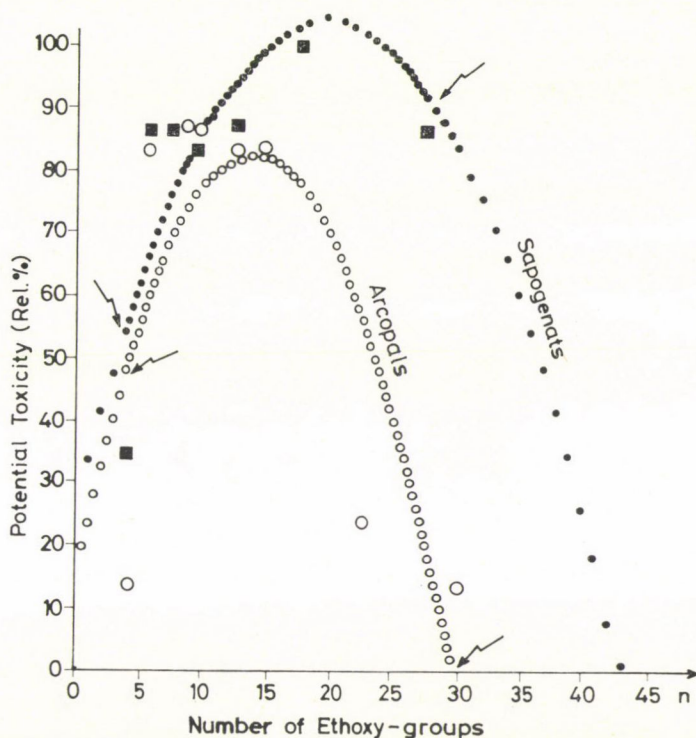


Fig. 2. Potential toxicity of nonionic tensides to tobacco leaves. Curves were drawn according to equations as follows:  $Y_A = 14.431 + 9.585 X_i - 0.338 X_i^2$  ( $r_A = 0.79 > r_{P=5\%} = 0.66$ ;  $n = 8$ );  $Y_S = 26.148 + 7.821 X_i - 0.196 X_i^2$  ( $r_S = 0.79 > r_{P=5\%} = 0.71$ ;  $n = 7$ ) where  $X_i$  is the number of ethoxy-groups in polyethyleneglycole chain of both *arcopals* ( $Y_A$ ) and *sapogenats* ( $Y_S$ ) labeled with symbols  $\circ$  and  $\blacksquare$ , respectively. The approximated values deviated from measured ones in the section of curves limited with arrows by 21.6 ( $Y_A$ ) and 12.9 ( $Y_S$ ) R% as averages

tributyl-phenyl series (arcopals and sapogenats) with medium length of ethylene oxide chains (Fig. 2) and the same was found with oleyl-alcohol series (genapols). Tweens exhibited medium toxicity with smaller variation by the age of leaves than did the other tensides.

The data concerning the phytotoxicity of tenside : cyclodextrin mixtures are compiled in Table 2. Cyclodextrins can prevent in some cases and at various extent the toxic action of tensides, depending on their molar ratio in the mixture.

The sensitivity of test plants to tenside *IV* increased in the order of tobacco, sugar-beet and spiderwort, and this order was not influenced by the various  $\beta$ -CD : tenside ratios (Table 3).



Table 3

Influence of  $\beta$ -CD on the evolution of plant response\* to *p*-nonylphenyl-decaglycolate (IV)

Test plant <sup>a</sup>	Treatment <sup>b</sup>	Concentration of tenside <sup>c</sup> mM				
		2	4	6	8	10
<i>Nicotiana tabacum</i>	IV	—**	48	24	12	12
	IV + $\beta$ -CD***	—	—	24	12	12
<i>Beta vulgaris</i>	IV	—	48	24	24	24
	IV + $\beta$ -CD	—	—	—	24	24
<i>Tradescantia albiflora</i>	IV	—	—	72	24	24
	IV + $\beta$ -CD	—	—	—	—	—

\* = The evolution of plant response means the time (hours) required for the development of collapse.

\*\* = (—) no collapse developed at all.

\*\*\* =  $\beta$ -CD was added to the solutions of tenside at 15 mg/ml. The  $\beta$ -CD itself does not induce collapse.

The significance of effect was tested by *F* probe:  $F_{P=0.1\%} = 11.78 > F^a = 8.17 > >P_{=1\%} = 6.51$ ;  $F_{P=0.1\%} = 17.14 > F^b = 11.70 > F_{P=1\%} = 8.86$ ;  $F_{P=0.1\%} = 9.73 > F^c = 7.12 > F_{P=1\%} = 5.56$  (Sváb, 1981).

### Discussion

The age of the leaf tissue altered significantly the phytotoxic effect of tensides. This finding may be due to the fact that the cell membrane composition of older cells differs from that of the younger ones (McKersie et al., 1978; Lees and Thompson, 1980). Therefore the membrane damaging effect of tensides manifests differently, similar to the age dependent response of leaves to toxins and pathogens (Goodman, Király and Zaitlin, 1967).

As the variances in gravity of damage have depended on the concentration of an inducer (tenside), we assume the presence of a limited number of receptor sites in the cell membrane. The protective effect of various cyclodextrins can also been explained by a decrease in the number of free tenside molecules, which leads to decreased probability of the interaction between them and the receptor sites. The degree of prevention depends on the toxicity of free tenside (i.e. the affinity of receptor site to it) on the cyclodextrin: tenside molar ratio and on the strength of complex formation. However, this last parameter is strongly influenced by the sterical correspondance of the appropriate cyclodextrin + tenside pairs; the bulky hydrophobic part of tenside has to fit adequately into the cyclodextrin cavity. The various sterical parameters of cyclodextrins and tensides explain the different preventive effects observed. If the protection of receptor sites in the membrane would lead to the preventive effect of cyclodextrines, the sterical differences in tenside structures could not affect the interaction so markedly.

The finding that methylated beta-cyclodextrins exhibited a higher effect than did unmethylated, draws attention to the fact that not only the



dimensions of  $\beta$ -CD cavity (being theoretically identical in each  $\beta$ -CD derivate) but also other molecular characteristics influence the complex formation.

We assumed that the difference in sensitivity of test plants to the tensides can be related either to the dissimilar structure of their cell membrane (i.e. the position of the receptor site) or to the various importance of the counteraction between tenside and receptor site in metabolic consequences.

### References

- Baicu, T., Jilaveanu, A. (1977): The prevention of Tobacco Mosaic Virus infections in tomato and pepper by some chemical substances. *Acta Horticulturae*, **58**, 435–460.
- Babiker, A. G. T., Duncan, H. J. (1975): Penetration of bean leaves by asulam as influenced by adjuvants and humidity. *Pest. Sci.* **6**, 655–664.
- Clifford, D. R., Hislop, E. C. (1975): Surfactants for the control of apple mildew. *Pestic. Sci.* **6**, 409–418.
- Cserháti, T., Szőgyi, M., Győrfi, L. (1986): Interaction of some non-ionic tensides with dioleil phosphatidylcoline, studied by charge-transfer chromatography. *J. Chromatogr.*, **349**, 295–300.
- Dunleavy, P. J., Cobb, A. H., Pallett, K. E., Davies, L. G. (1982): The involvement of stomata in bentazol action in *Chenopodium album* L. Proc. 1982. *British Crop Prot. Conf. Weeds*, **1**, 187–191.
- Freed, V. H., Witt, J. M. (1969): Physicochemical principles in formulating pesticides relating to biological activity. *Adv. Chem. Ser.*, **86**, 70–80.
- Goodman, R. N., Király, Z., Zaitlin, M. (1976): *The biochemistry and physiology of infectious plant disease*. D. Van Norstrand Co. Inc, Princeton, New Jersey.
- Koch, J. (1982): *Stabilisation and controlled release of perfume in detergents*. Proc. 1<sup>st</sup> Int. Symp. on Cyclodextrins (ed: J. Szejtli), Akadémiai Kiadó, Budapest, 1982. p. 487–496.
- Králová, K., Mitterhauserová, L., Szejtli, J. (1983): Studium der Mizellbildungsprozesse von Tensiden des Typs N,N'-bis-(alkyldimethyl)-1,6-hexandiammoniumdibromid in wäßrigen Lösungen. II. Beeinflussung des Mizellbildungsprozesses durch Wechselwirkung mit  $\beta$ -Cyclodextrin. *Tenside Detergents* **20**, 37–39.
- Lees, G. L., Thompson, J. E. (1980): Lipid composition and molecular organization in plasma membrane-enriched fractions from senescing cotyledons. *Physiol. Plantarum* **49**, 215–221.
- Leroux, G. D., Harvey, R. G. (1981): Field evaluation of grass herbicides for quack grass control in alfalfa. *Proc. North Central Weed Control Conf.* **36**, 40–41.
- Lewi, P. J. (1976): Spectral mapping, a technique for classifying biological activity profiles of chemical compounds. *Arzneimittel-Forschung*, **26**, 1295–1300.
- McKersie, B. D., Lepock, J. R., Knuuv, J., Thompson, J. E. (1978): The effects of cotyledon senescence on the composition and physical properties of membrane lipid. *Biochim. Biophys. Acta* **508**, 197–212.
- Müller, R., Burth, U. (1983): Benzimidazol-Tenzid-Formulierung mit Wirksamkeit bei Benomylresistenz. *Abhandl. Akad. Wiss. DDR*, N1 1982. Nr. 1. 315–319.
- Okubo, T., Kitano, H., Ise, N. (1976): Conductometric studies on association of cyclodextrin with colloidal electrolytes. *J. Phys. Chem.*, **80**, 2661–2664.
- Parochetti, J. V., Wilson, H. D., Beste, C. A. (1977): Effect of several adjuvants on pre-emergence and postemergence herbicides in 1976. *Proc. of the Northeastern Weed Sci. Soc. Baltimore*, **31**, 105–112.
- Spotts, R. A., Peters, B. B. (1982): Use of surfactants with chlorine to improve pear decay control. *Plant Disease*, **6**, 725–727.
- Sváb, J. (1981): *Biometrical Methods in Research Work*. Mezőgazdasági Kiadó, Budapest, 1981.
- Szejtli, J. (1982): *Cyclodextrins and their Inclusion Complexes*. Akadémiai Kiadó, Budapest.
- Szejtli, J. (1984): *Industrial applications of cyclodextrins*. In: *Inclusion Compounds* (Eds: Atwood, J. L. Davies, J. E. and McNicol, D. D.) Vol. III. Academic Press, London, 1984, 331–338.
- Szőgyi, M., Tölgyesi, F., Cserháti, T. (1983): *Correlation between ion permeability and structure of model membranes modified by tensides*. In: *Physical Chemistry of Transmembrane Ion Motions* (Ed.: Spach, D), Elsevier Sci. Publishers B. V. Amsterdam, 1983. 29–36.
- Van Valkenburg, W. (1973): *Pesticide Formulations*. Marcel Dekker Inc., New York, 1973.





## ANTAGONISM OF MAGNESIUM AND ALUMINIUM IN BEAN AND WHEAT

S. A. KISS

BORSOD CHEMICAL WORKS, KAZINCBARCIKA, HUNGARY

(Received 26 August 1987; accepted 8 December 1987)

Aluminium has a toxic effect primarily on the roots of plants, which become deformed and show a coralloid growth habit; and it reduces the ability of roots to take up nutrients in general, and magnesium in particular which thereby lessens the plant yield. The increasing magnesium content of the nutrient solution gradually hinders the aluminium uptake and thus decreases the toxicity. According to our investigations, aluminium and magnesium are mutual antagonists. Tolerance requires the ratio of Mg: Al to be above two in the nutrient solution. The toxicity of aluminium is species- and variety dependent as demonstrated on bean and wheat plants. Aluminium also damages the *Rhizobium*, but magnesium increases the tolerance in this case too.

**Keywords:** aluminium, antagonism, bean, magnesium, *Rhizobium*, wheat

### Introduction

The acidification of our soil and water has become an increasing problem which currently concerns soil science, plant physiology, crop production and even human biology. The acidification of soils and the resulting aluminium toxicity are not new problems. In his book published in 1928 Kreybig wrote about these phenomena and the "harmful mobile aluminium" thus released, and suggested liming to eliminate them.

According to Runge (1984) it is the release of various materials, primarily of the toxic  $\text{Al}^{3+}$ -ion, rather than the pH of the soil solution, that damages plants. The aluminium content of soils, about 4% is rather high. Aluminium is mostly found in the clay-mineral soils, as water-insoluble aluminium silicate, which is not toxic. Only the dissolved ionic aluminium causes damages. In the soil solution first of all the  $\text{Al}^{3+}$ -ion, and occasionally its monomeric hydrolysis products — the  $\text{Al}(\text{OH})^{2+}$ - and  $\text{Al}(\text{OH})^{+}$ -ions, are present. In acid soils the quantity of soluble ionic aluminium greatly increases. The solution of aluminium is pH-dependent. Gahamani (1977), carrying out examinations with terra rossa, found that when the pH-value of the soil fell from 5.5 to 4.5 the  $\text{Al}^{3+}$ -ion concentration of the soil solutions grew sixfold.

Aluminium is decidedly a root toxin, as the first symptoms always appear on the roots, which then exhibit a so-called "coralloid" growth habit. This comes about when the apical meristem necrotizes and, as new branch



roots develop, their tips also soon necrotize so that, with the short, blunt branches produced, the root-system takes on the appearance of a corall growth (Runge 1984). This malformation of the roots inhibits the nutrient supply, as well as decreasing the water- and nutrient transporting capacity of the vascular bundles in the shoots. The blocking of vascular bundles causes particularly great damages — even a sudden decay of trees — in forests. Aluminium toxication therefore produces nutrient deficiency symptoms, even when the soil contains sufficient nutrients (Guerrier 1979). The Ca- and Mg uptake decreases in a particularly great measure in response to toxication by aluminium.

Aluminium toxication damages the DNA- (Wallace and Anderson 1984) and RNA (Matsumoto and Morimura 1980) synthesis and the photosynthesis (Sarkuman 1984).

According to Westendorf (1982) the  $\text{Ca}^{2+}$ -ions prevent the aluminium-uptake and its harmful effect.

Starting from the above data, we examined the question of whether the  $\text{Mg}^{2+}$ -ions would also hinder the uptake of harmful aluminium ions. The present paper describes our experiments in regard to the Al-Mg antagonism.

### Materials and methods

In our experiments we used bean- and wheat plants as indicators, in modified Prjanyisnyikov perlite culture fluid. The modification concerned the Mg content (1.25; 2.50; 5.00 mmol) and the Al supplement (0.02; 0.19; 1.80 mmol). The pH of the solution was adjusted to 4.5. The water lost by the culture pots in the glasshouse was replaced every day by "ion-free" water of pH 4.5, and 100 ml from the culture fluid was added every week, in order to ensure the nutrient supply that was determined by the development of the control plants. There were 6 replications, and the treatments are shown in Table 1.

Table 1  
*Pearlite culture pot experiments, variations and labels  
for Al and Mg*

Serial number of treatment	Al mmol/L	Mg mmol/L
1	0	1.25
2	0.02	1.25
3	0.19	1.25
4	1.80	1.25
5	0	2.50
6	0.02	2.50
7	0.19	2.50
8	0.80	2.50
9	0	5.00
10	0.02	5.00
11	0.19	5.00
12	1.80	5.00

Owing to the variety dependence of aluminium sensitivity (Klimashewsky 1972) our experiments were performed with 4 green (dwarf) bean varieties ("Valja", "Róna", "Cherokee" and "Budai piacos") and 1 wheat variety ("Jubilejnaja"). We chose these two plant species because of the aluminium sensitivity of wheat as a monocotyledonous- and of bean as a dicotyledonous plant. Thus, in case the favourable effect of magnesium for tolerance to aluminium toxicity be proven true for both plants, then the phenomenon could be considered valid in a wider range of plants.

The bean seeds were given *Rhizobium* treatment (inoculation with a mixture of the strains B-1, B-3 and Friol) in order to examine the Al-Mg effect on nodule formation. The *Rhizobium* inoculum was obtained from the Research Institute for Soil Science and Agrochemistry for which we here express our thanks.

### *Morphological examinations*

The development of the plants was followed visually as well as by weighing the shoots and roots. In the case of bean flower, the pod and nodule formations were also taken into consideration. During the examinations photographs were taken of the plant parts examined. For bean the nodules were both counted and weighed. In addition to the shape of roots, their coralloid habit was also examined.

### *Aluminium uptake*

The aluminium uptake by the roots was examined in three ways:

(a) The cations have a definite positive effect (proportionate to their concentration and charge in the root) on the anion uptake of roots, which can be measured by the absorption of a 0.002 mol solution of acidic indigo carmine (semiquantitative method, Koloszov 1948).

(b) The aluminium ions form red-violet "lacquer" with quinalizarin, so — according to Kalovoulos and Misopolinos (1983) — they can be observed in the roots (semiquantitative method). This renders it possible to examine in root sections the location (penetration) of aluminium. For the examination, 100 mg quinalizarin was dissolved in 20 ml pyridine, then diluted with acetone to 200 ml. Freshly cut roots were washed in ion-free water. Then the above reagent was applied by drops on the end or section of each root and let run down to the root tip. The roots (sections) were held for one minute above a glass vessel containing concentrated  $\text{NH}_4\text{OH}$ , then placed above acidum aceticum for another minute. The red-violet colour indicated the place where the aluminium ions were located.

(c) After the careful reduction of the samples (shoot, root) to ashes and the hydrochloric absorption of the ash, the Mg- and Al content was determined by atom absorption spectrophotometry.

## **Results**

### *Results of measurements*

The plant length- and shoot-, root- and nodule weight data obtained for the bean varieties are listed in Table 2, 3, 4 and 5. The measuring results show that, while a small quantity of aluminium increased the yield to a varying extent depending on the variety, higher aluminium concentrations (1.8 mmol) caused in each case depression and coralloid root formation.

Figures 1 and 2 clearly show the difference in green mass and root between plants of the variety "Róna" grown in aluminium-free medium (1) and those raised in a medium containing 1.8 mmol aluminium (4). The former (1) possessed abundant rootlets and nodules on the roots, while in treatment (4) there were no nodules and hardly any rootlets (coralloid roots). With the variety "Valja" there were a few nodules and some rootlets in the 1.8 mmol aluminium treatment. In the case of the variety "Cherokee" the rootlet- and



nodule formation, while decreasing with the aluminium concentration, was intensive in each treatment; that is, this variety was tolerant to aluminium toxication. The toxicity of aluminium was best tolerated by the variety "Budai piacos" (Table 5). The maximum damage observed with this variety was 10% for shoots and 25% for roots. Full tolerance could probably be

Table 2

*Results of experiments with the dwarf bean variety "Róna"  
treated with various rates of Al and Mg*

Serial number	Treatment mmol/l		Length of plant		Flower + pod		Green mass		Root weight		Nodule weight	
	Al	Mg	cm	%	n/plant	%	g/plant	%	g/plant	%	g/plant	%
1	0	1.25	35.08	100	6.49	100	14.30	100	5.25	100	0.66	100
2	0.02		33.02	94	3.87	60	10.25	71	3.03	58	0.32	48
3	0.19		28.45	81	3.74	58	9.34	65	2.17	41	0.15	22
4	1.8		23.56	67	1.00	15	4.03	28	1.46	28	0.0	0
5	0	2.50	36.13	103	6.72	103	15.02	105	5.31	101	0.70	106
6	0.02		31.51	90	5.07	78	9.62	67	2.21	42	0.38	57
7	0.19		27.90	79	3.68	57	9.86	69	2.10	40	0.11	16
8	1.80		25.07	71	1.20	18	5.28	36	1.48	28	0.0	0
9	0	5.00	39.9	114	6.60	101	14.40	100	5.20	99	0.51	77
10	0.02		33.28	95	5.28	81	10.26	72	2.42	46	0.25	38
11	0.19		30.08	85	4.15	64	12.10	85	2.22	42	0.16	24
12	1.80		26.31	75	1.42	22	6.11	43	1.60	30	0.0	0

Table 3

*Experiment data of the dwarf bean variety "Valja" treated with various rates of Al and Mg*

Serial number	Treatment mmol/l		Length of plant		Flower + pod		Green mass		Root weight		Nodule weight	
	Al	Mg	cm	%	n/plant	%	g/plant	%	g/plant	%	g/plant	%
1	0	1.25	29.29	100	6.56	100	9.56	100	3.44	100	0.83	100
2	0.02		31.56	107	6.50	99	10.04	105	3.22	93	0.72	87
3	0.19		30.33	103	7.16	109	7.37	77	3.03	88	0.60	72
4	1.80		20.34	69	0.54	8	2.44	25	0.88	26	0.0	0
5	0	2.50	29.00	99	6.16	93	9.76	102	3.65	106	1.05	126
6	0.02		30.57	104	8.11	123	10.97	114	3.43	100	0.97	117
7	0.19		27.93	95	5.00	76	8.12	85	3.12	90	0.62	75
8	1.80		24.6	84	3.18	48	3.66	38	1.65	48	0.01	1
9	0	5.00	28.70	98	5.75	88	9.46	99	3.02	88	0.51	61
10	0.02		28.82	99	5.98	91	10.48	109	1.62	47	0.32	38
11	0.19		27.90	95	5.66	86	7.57	79	1.43	41	0.18	21
12	1.80		25.02	85	4.24	64	6.41	67	1.24	36	0.01	1

Table 4

*Experiment data of the dwarf bean variety "Cherokee" treated with Al and Mg*

Serial number	Treatment, mmol/lit		Length of plant		Green mass		Root weight		Nodule weight	
	Al	Mg	cm	%	g/plant	%	g/plant	%	g/plant	%
1	0	1.25	27.2	100	5.5	100	3.2	100	0.52	100
2	0.02		28.2	104	5.8	105	3.6	112	0.48	92
3	0.19		27.5	101	5.2	94	3.4	106	0.42	80
4	1.80		22.3	82	4.2	76	3.5	109	0.25	48
5	0	2.50	27.6	102	6.5	118	4.0	125	0.60	115
6	0.02		25.6	98	5.8	105	3.8	118	0.39	75
7	0.19		25.3	93	5.4	98	3.0	93	0.26	50
8	1.80		23.4	86	4.7	85	3.0	93	0.30	58
9	0	5.0	28.3	104	7.0	127	3.6	112	0.31	59
10	0.02		29.4	108	5.8	105	2.9	90	0.28	54
11	0.19		24.3	89	5.8	105	2.2	69	0.23	44
12	1.80		23.6	87	5.5	100	2.3	71	0.18	35

Table 5

*Experiment data of the dwarf bean variety "Budai piacos" treated with Al and Mg*

Serial number	Treatment, mmol/lit		Length of plant		Green mass		Root weight		Nodule weight	
	Al	Mg	cm	%	g/plant	%	g/plant	%	g/plant	%
1	0	1.25	21.9	100	4.59	100	3.40	100	0.048	100
2	0.02		23.1	106	4.35	95	3.30	97	0.040	83
3	0.19		23.0	105	4.50	98	2.90	85	0.032	56
4	1.80		21.3	97	4.14	90	2.57	75	0.020	42
5	0	2.50	23.8	109	5.79	126	3.52	103	0.085	177
6	0.02		22.7	104	5.74	125	2.98	87	0.052	108
7	0.19		23.0	105	5.34	116	3.12	92	0.035	73
8	1.80		20.8	95	4.97	108	2.61	76	0.026	54
9	0	5.00	22.1	101	6.66	145	4.63	136	0.140	291
10	0.02		23.7	108	7.76	169	4.09	120	0.090	187
11	0.19		23.7	108	6.46	140	3.83	113	0.051	106
12	1.80		19.5	89	5.10	111	3.40	100	0.029	60

achieved with the magnesium treatment (treatment 12.) because this variety was responsive to magnesium.

The bean varieties showed the following order of sensitivity to aluminium toxicity:

"Róna" > "Valja" > "Cherokee" > "Budai piacos"



Figure 3 demonstrates the favourable effect of the fourfold (5 mmol) magnesium concentration in treatment 12., compared to treatment 4. In which the aluminium concentration was the same (1.8 mmol) but the magnesium concentration was lower (1.25 mmol).

However, the increasing magnesium supply counterbalanced, to some extent, the inhibition caused by aluminium in the sensitive varieties, but



Fig. 1. Inhibitory effect of aluminium on "Róna" beans. (1) control, without Al, in the case of 1.25 mmol Mg, (4) 1.8 mmol Al and 1.25 mmol Mg



Fig. 2. Damage done by aluminium to the roots of "Róna" beans. (1) control, without Al, with 1.25 mmol Mg, (4) 1.8 mmol Al and 1.25 mmol Mg



Fig. 3. The harmful effect of aluminium decreased by magnesium in the bean variety "Róna".  
(4) 1.8 mmol Al and 1.25 mmol Mg, (12) 1.8 mmol Al and 5.00 mmol Mg

could not prevent it even at a 2.5-fold concentration. In the tolerant varieties the inhibition could be perfectly relieved. The 5 mmol magnesium concentration often had a depressive effect on the aluminium sensitive varieties, although they fully tolerated the 1.8 mmol aluminium concentration. Sometimes a small quantity of aluminium even had a stimulatory effect.

The results of wheat experiments are seen in Table 6 judging from the data, wheat (variety: "Jubilejnaja") is less sensitive to aluminium toxicity than some of the bean varieties. Magnesium relieved (in some cases eliminated) the inhibition by aluminium in wheat too (Fig. 4). High concentrations of magnesium, as in the case of bean, had a depressive effect.

Table 6

*Measuring data of a culture pot experiment with the wheat variety "Jubilejnaja" treated with various rates of Al and Mg at the age of 2 months*

Serial number	Treatment, mmol/lit		Length of plant		Green mass		Root weight	
	Al	mg	cm	%	mg/plant	%	mg/plant	%
1	0	1.25	31.8	100	112	100	35	100
2	0.02		27.9	88	120	107	50	142
3	0.19		28.8	90	88	78	40	114
4	1.8		13.2	41	25	22	14	40
5	0	2.50	29.5	92	120	107	38	108
6	0.02		27.6	88	107	95	36	103
7	0.19		27.9	88	100	85	36	103
8	1.80		16.5	52	33	29	22	62
9	0	5.00	27.4	86	107	96	32	91
10	0.02		26.5	83	103	92	39	111
11	0.19		23.1	73	95	85	29	83
12	1.80		16.8	53	41	36	36	100



### Results of aluminium uptake

(a) According to the results of root examinations with indigo carmine the stain uptake (the tint) of roots as a function of the aluminium concentration can be described by a maximum curve. The extent of stain uptake: was  $\emptyset \text{Al} \ll 0.02 \text{ Al} < 0.19 \text{ Al} > 1.85 \text{ Al}$ . The double- and fourfold magnesium



Fig. 4. The harmful effect of aluminium decreased by magnesium in wheat

concentration decreased the shade of colour, i.e. the uptake of stain, in plants raised in culture fluids of identical aluminium content. This suggests that the magnesium ions inhibit the aluminium uptake by plants.

(b) With the quinalizarine test we wished to find out whether the aluminium ions are only adsorbed into the surface cells or else penetrate to some depth into the roots. From the main roots of bean plants raised in culture fluids with different aluminium- and magnesium concentrations, sections were made and stained with quinalizarine. It was found that only the outer part of the root (epidermis, hypodermis, endodermis) showed intensive staining, while its inner part remained unchanged. This agrees with the results obtained by Wagatsuma (1984); namely, that the main obstacle to aluminium translocation must be in the endodermis. Under the influence of an increasing magnesium concentration, the aluminium ions become less and less able to penetrate into the inside of the root (indicated by the poor colouring). This is due to the inhibitory effect of magnesium on aluminium uptake.

(c) The results of aluminium- and magnesium determination by atom absorption spectrophotometry are seen in Table 7. The aluminium concentration of the root agrees with that observed with the stain anion uptake; that is, the aluminium uptake can be described with a maximum curve and the



Table 7

*Effect of Al- and Mg treatments on the Al- and Mg content in the shoot and root of bean (mg/g dry matter)*

Serial number	Treatment, mmol/lit.		Mg		Al	
	Al	Mg	Shoot	Root	Shoot	Root
1	0	1.25	2.42	4.89	0.006	0.016
2	0.02		2.68	6.03	0.010	0.023
3	0.19		2.30	4.20	0.005	0.601
4	1.80		2.40	1.80	0.006	0.441
5	0	2.50	2.85	9.18	0.006	0.010
8	1.80		2.60	4.03	0.005	0.330
9	0	5.00	4.36	5.86	0.005	0.005
12	1.8		3.72	3.20	0.011	0.092

magnesium inhibits the aluminium uptake. In the shoots the aluminium content increased at a lower rate than in the roots. This corresponds to the literary data of experiments carried out with other plants.

### Conclusions

The results of our experiments show that the inhibition of root- and shoot growth caused by aluminium is a variety-dependent feature in the case of beans. Increasing concentrations of magnesium counterbalance, to some extent, the harmful effect of aluminium. The extent to which magnesium makes the plants tolerant to aluminium toxicity also depends on the variety. In varieties with higher tolerance, the favourable effect of magnesium is stronger.

The sensitivity of *Rhizobium* to aluminium toxicity was studied by Mayz de Manzi (1964) and Wood (1984). Both state that there are aluminium-tolerant and aluminium-sensitive strains although. They only examined plants belonging to the family *Fabaceae* (crimson clover). According to the results of our experiments, the nodule formation depends not only on the aluminium sensitivity of the *Rhizobium* strain(s), but is also closely related to the aluminium tolerance of the host plant. In a tolerant variety, the nodule formation is much more intensive than in a sensitive variety with the same concentration of aluminium. This can be explained by the poor nutrient supply in the roots of aluminium-sensitive plants compared to tolerant plants, in the roots of which the nutrient supply remains next to optimum in the case of aluminium toxication.

Growing concentrations of aluminium increase the aluminium fixation and, hence the uptake of anions ( $\text{PO}_4^-$ , stain anion) only to a certain limit; because a high aluminium concentration impairs the adsorption capacity of



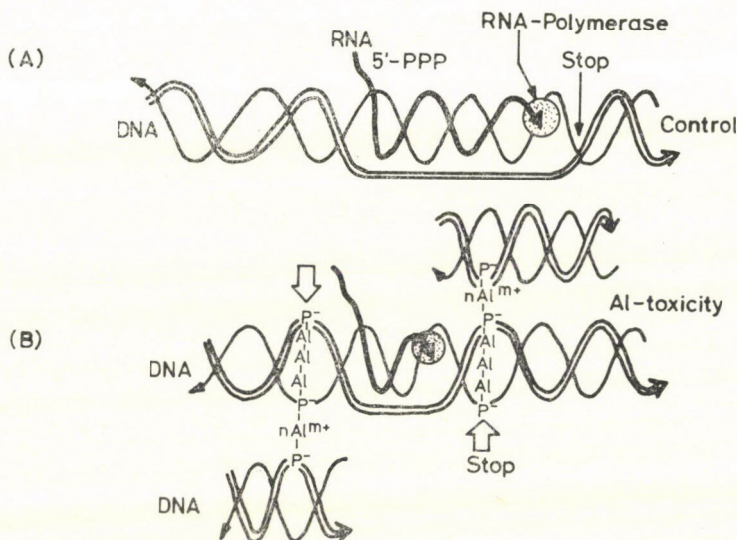


Fig. 5. Supposed mechanism of transscription inhibition by aluminium (Matsumoto 1980)

the root and a smaller quantity of aluminium will be fixed (Lee 1984). According to Klimashevski (1976) large quantities of aluminium entering the cell wall decrease the membrane permeability. Deleers (1986) explains this phenomenon with the membrane rigidity which occurs already at a  $25 \mu m$  concentration of aluminium. The reduced membrane permeability is responsible for the poor uptake of nutritive elements (e.g. Ca, Mg, Lee 1984), and for this reason the plant remains underdeveloped. Since the magnesium inhibits the adsorption of aluminium and its penetration into the root cells, the permeability of the cell wall suffers only slight damage, any. The same is reported of the calcium ions by Wagatsuma (1983) and Westendorf (1982).

Aluminium inhibits the synthesis of DNA and RNA by linking into the double spiral of DNA (Fig. 5) to the phosphates, forming as it were a blockage of transcription (Matsumoto and Morimura 1980), so that the synthesis of DNA and RNA stops. Magnesium is also linked with the phosphates in the DNA and increases the stability of the latter (Eichhorn 1968), thereby preventing aluminium from entering. This too may be an explanation for the inhibitory effect of magnesium on aluminium toxicity.

It should be noted that a small quantity of aluminium may even have a stimulatory effect, and that magnesium is only able to prevent toxication by aluminium when the ratio of Mg : Al in the culture fluid is above 2. Otherwise, a too high concentration of magnesium may adversely affect the growth of both plant and nodules — again depending on the variety. Accordingly, antagonism and synergism depend on the concentration and on the ratio of the two ions alike.



On the basis of our measurements it can be established that the magnesium and aluminium are mutual antagonists, therefore a better supply of magnesium decreases the harmful effect of aluminium. This agrees with the observation made by Guerrier (1979), that aluminium inhibits magnesium uptake. Thus, dolomite (with its magnesium content) is — in our opinion — a better meliorative than lime stone for acidic soils (of high H- and Al-ion content).

### References

- Deleers, M., Servais, J. P., Wülfert, E. (1986): Neurotoxic cations induce membrane rigidification and membrane fusion at micromolar concentrations. *Biochim. Biophysic. Acta*, **855**, 271–276.
- Eichhorn, G. L., Shin, Y. A. (1968): Interaction of metal ions with polynucleotides and related compounds. XII. The relative effects of various metal ions on DNA heredity. *J. A. Chem. Soc.*, **90**, 7323–7330.
- Gahamani, A. B. (1977): Eksztrahirnemij i obmeniji aljuminij v iskusztvennih preparatah mineral szubtropicseszkijh pocsvah. *Pocsvedenie*, **12**, (11) 43–53.
- Guerrier, G. (1979): Adsorption des éléments minéraux en présence d'Aluminium. *Plant Soil*, **51**, 275–278.
- Kalavoulos, J. M., Misopolinos, N. D. (1983): Aluminium detection on corn roots by the quinallizarin method. *Plant Soil*, **74**, 131–132.
- Klimashewsky, E. L., Markova, Yu. A., Bernatzkaya, M. L., Malysheva, A. S. (1972): Physiological responses to Al toxicity in root zone of pea varieties. *Agrochimica*, **16**, 487–496.
- Kolosov, I. I. (1948): Vlijanie kationov na pogloshenie kisljüh kraszok i anionov mineralnüh szolej kornjani rasztenij. *Trudü Inszt. Fiziol. Raszt.*, A. N. USSR, **6**, 1.
- Kreybig, L. (1928): *A talaj élete, javítása és trágyázása* (Life, melioration and fertilization of the soil). Budapest.
- Lee, J., Pritchard, M. W. (1984): Aluminium toxicity expression on nutrient uptake, growth and root morphology of *Trifolium repens* L. *Plant Soil*, **82**, 101–116.
- Matsumoto, H., Morimura, S. (1980): Repressed template activity of cromatin of pea roots treated aluminium. *Plant Cell Physiol.*, **21**, 951–959.
- Mayz de Manzi, J., Cartwright, P. M. (1984): The effects of pH and Al toxicity on the growth and symbiotic development of cowpeas. *Plant Soil*, **80**, 423–430.
- Runge, M. (1984): Bedeutung von Aluminium als Standortfaktor. *Düsseldorfer Geobot. Kolloquien*, **1**, 30.
- Saigusa, M., Shoji, S., Takahashi, T. (1980): Plant root growth in acid andosols from North-eastern Japan. *Soil Sci.*, **130**, 242–250.
- Sarkunan, V., Biddappa, C. C., Nayak, S. K. (1984): Physiology of Al toxicity in rice. *Current Sci. India*, **53**, 882–884.
- Wagatsuma, T. (1983): Effect of nonmetabolic conditions on the uptake of Al by plant roots. *Soil Sci. Plant Nutr. (Tokyo)*, **29**, 323–333.
- Wagatsuma, T. (1984): Characteristics of upward translocation of Al in plants. *Soil Sci. Plant Nutr. (Tokyo)*, **30**, 345–358.
- Wallace, S. K., Anderson, I. C. (1984): Aluminium toxicity and DNA synthesis in weath roots. *Agron. J.*, **76**, 5–8.
- Wood, M., Cooper, J. E., Holding, A. J. (1984): Aluminium toxicity and nodulation of *Trifolium repens*. *Plant Soil*, **78**, 381–391.





## PURIFICATION AND CHARACTERIZATION OF MYOSIN FROM ISOLATED CHLOROPLASTS OF SPINACH LEAVES (*SPINACEA* *OLERACEA* L.)

R. NEHÉZ,<sup>1</sup> S. FAZEKAS,<sup>2</sup> É. SÁRVÁRI<sup>3</sup>, I. ÓVÁRY<sup>4</sup> and  
V. SZÉKESSY-HERMANN<sup>2</sup>

<sup>1</sup>CEREAL RESEARCH INSTITUTE, SZEGED, HUNGARY

<sup>2</sup>2nd INSTITUTE OF BIOCHEMISTRY AND <sup>4</sup>PSYCHIATRIC CLINIC,  
SEMMELWEIS UNIVERSITY MEDICAL SCHOOL, BUDAPEST, HUNGARY

<sup>3</sup>DEPARTMENT OF PLANT PHYSIOLOGY, LORÁND EÖTVÖS UNIVERSITY,  
BUDAPEST, HUNGARY

(Received 1 September 1987; accepted 15 February 1988)

Myosin was prepared from the chloroplast fraction of spinach leaves. The myosin content of the chloroplast comprises about 1.6–2% of the total protein. The myosin preparations were prepared with a significant endogenous phosphate content (24–34 mol P/mol), which binds to different basic amino acids to form N-P type phosphoryl groups. The fresh preparations of chloroplast myosin could be phosphorylated to a higher saturation degree, and they have the ability to form filamentous aggregates.

**Keywords:** Characterization, myosin, purification, spinach, *Spinacea oleracea*

### Introduction

It was reported more than 20 years ago (Onishi 1964) that contractile proteins occur in chloroplasts. Later authors are of the opinion that the contractile fibrils are responsible for the active mechanical motion and changes in volume of the chloroplast (Schorer-Mörtel 1972, Schönboom, 1973), and also that they take part in the endogenous particular motion of the chloroplast and in transport processes (Kroon 1969, Tandler and Hoppel 1972, Sokolov et al. 1986). A few authors (Schaitman and Greenwalt 1968, Ster and Palmer 1984, Briat 1986) consider that the contractile proteins of the main organelles (chloroplast, mitochondrion) must be sought among the structural elements of their matrix.

The flow motion of the chloroplasts and mitochondria in the cytoplasm of plant cells, and even the motion in the nucleus and membranes, is attributed to the contractile system (Wagner et al. 1972, Williamson 1976, Menzel and Schliwa 1986). Nehéz et al. (1980, 1985) obtained a considerable amount of myosin from meristematic tissues, i.e. from the root growing points of sprouting maize, whose cells have only a primary cell wall.

On the basis of published data and our own experience, we believe that plant cells, like the cells of the liver parenchyma, contain two types of myosin



with different functions: particular myosin (chloroplast, mitochondrium) and cytoplasmic myosin (Fazekas et al. 1987). In the present work an appreciable quantity of chloroplast has been prepared, with a view to characterizing the properties of this chloroplast myosin and comparing them with those of myosin obtained from other sources.

### Materials and methods

All chemicals used were commercially available products of analytical grade unless otherwise stated: ammonium molybdate,  $H_2SO_4$ ,  $HNO_3$  (Merck, Darmstadt), 2-mercapto-ethanol, DEAE-cellulose, TRIS (tris-hydroxymethyl-aminomethane), Dextran blue (Serva, Heidelberg), DTT (dithiothreitol), Triton X-100 (Calbiochem), Sepharose CL 4B (Pharmacia, Uppsala), EGTA (ethylene glycol bis/2-aminoethylether/-N,N'-tetraacetic acid) (Fisons, England), EDTA (ethylene diamine tetraacetic acid), methanol, ethanol, acetone, KCl,  $KHCO_3$ , KOH, NaCl,  $MgCl_2$ , ATP (Reanal, Budapest).

The chloroplast preparation was obtained from spinach leaves by the method of Walker (1980), with 0.33 M saccharose solution containing 1 mM  $MgCl_2$ . In general, 50–62 cm<sup>3</sup> (but on repetition of the final step only 40–50 cm<sup>3</sup>) chloroplast past can be obtained from 1 kg freshly collected and cleaned leaves. The paste contains about 1700–1900 mg total protein.

Due to the high chlorophyll content, the preparation of myosin from chloroplasts required a more careful purification procedure than the preparation of other myosins of plant origin from chlorophyll-free tissues, as detailed earlier (Nehéz et al. 1985).

The chloroplast paste was cooled to 0 °C and uniformly mixed with 1 ml 2-mercapto-ethanol (added dropwise). The product was then mixed with the same volume of 1.2 M KCl solution containing 8 mM  $KHCO_3$ , and the mixture was homogenized at 1500 rpm for  $4 \times 10$  s in a 50 ml Potter-Elvehjen homogenizer, furnished with a motor-driven teflon pestle, and then centrifuged at 15.000 g for 10 min. The supernatants were collected in a dialysis tubing and dialysed against distilled water. The chloroplast sediment was rehomogenized in 35 ml 0.6 M KCl + 8 mM  $KHCO_3$  solution (pH 9.3), and left to stand overnight in a refrigerator in order for further myosin to be extracted. The following morning, this mixture was centrifuged as above, and the pellet fraction again extracted with 3–5 ml 2 M KCl + 8 mM  $KHCO_3$  solution and centrifuged after 1 h. The extracted residue was used for the preparation of Straub actin. The supernatants were combined in a dialysis tubing and dialysed against distilled water until the disappearance of myosin floccules (10–15-fold volume, 4 changes).

The flocculated myosin was collected by centrifugation at 15.000 g for 20 min. The pH of the supernatant was adjusted to 7.6 with  $KHCO_3$  solution, and then mixed with 10 mg fibrous DEAE-cellulose suspension for adsorption of the myosin remaining in the supernatant and its recovery by centrifugation. The pellet fractions were solubilized by mixing with a little 2 M KCl +  $KHCO_3$  solution (pH 9.3), then combined; and the KCl concentration was adjusted to 0.6 M by dilution. Further dilution was then done with 0.6 M KCl +  $KHCO_3$  solution, so that the protein concentration should decrease to about 10 mg/ml. The cloudy solution was clarified by centrifugation at 5000 g for 20 min. The pellet fraction was reextracted with 3–5 ml 0.6 M KCl +  $KHCO_3$  solution, followed by centrifugation and combination with the main fraction.

The KCl concentration of the supernatant was adjusted to 0.38 M, and its pH to 7.6; and the supernatant was mixed with 5 g fibrous DEAE-cellulose suspension and centrifuged as above. By means of the adsorption, a considerable amount of RNA and phospholipid was removed from the myosin solution.

The myosin was again flocculated by dialysis, and collected by centrifugation, as above. The pellet fraction was mixed with 1–2 ml 2 mM DTT solution for at least 10 min under ice-cooling, and the myosin was redissolved by mixing with small volumes of 0.6 M KCl +  $KHCO_3$  solution. Efforts were made to obtain a myosin solution in the smallest possible volume (6–10 ml).

If the myosin solution was still cloudy, it was clarified by centrifugation at 5000 g for 20 min., but if it was only mildly opalescent and a shade of green, the preliminary centrifugation was omitted and it was mixed with 2–3 mg DEAE-cellulose suspension and ultracentrifuged at 105.000 g for 120 min. to remove the denatured proteins, actomyosin, and RNA and phospholipid traces (uc-myosin). Finally the ultracentrifuged myosin was purified by gel filtration



on a Sepharose CL 4B column. The myosin was contained in the tubes of the first protein fraction peak.

At the used size and content of column, the myosin was received at given ml-s and the mean for myosin was somewhat above 470.000 Dalton). Data for myosin were calculated for 478.000 D as a probable molecular mass.

Protein and phosphate (P) content determination, the investigation of phosphorylation ability, and the removal of superfluous nucleotide and total lipid from the samples were performed as detailed in (Fazekas et al. 1981, Nehéz et al. 1985, Nehéz et al. 1986) for separation of the P-containing fraction, the alkaline hydrolysates of lipid-free myosins were used. Hydrolysis was carried out exclusively in 10 ml teflon ampoules (furnished with teflon screw caps) in the presence of 3 M KOH, at 105 °C 10 h. By ion-exchange chromatography, the P-containing fractions of the hydrolysates could be resolved into 7–9 peaks (see Figure 2).

## Results

50–60 ccm chloroplast paste could be obtained from 1 kg leaves containing 1700–1900 mg total protein. The isolation procedure yielded 15–17 mg gel-filtered myosin from 1 kg leaves, which corresponded to about 1% of the chloroplast total protein. The loss in the preparation of myosin in this procedure was assessed as about 45–50%, and thus the total myosin concentration

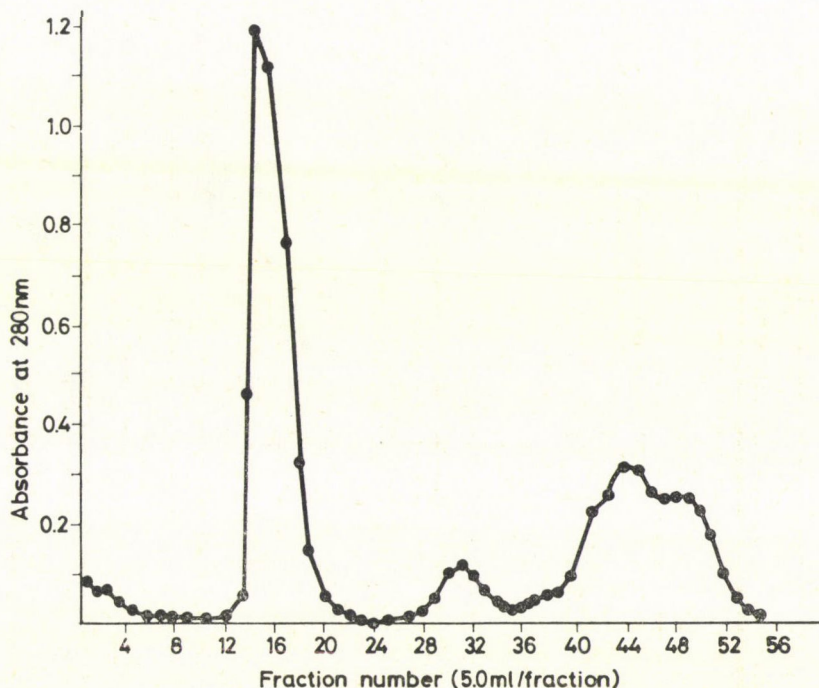


Fig. 1. Gel filtration and elution profile of ultracentrifuged chloroplastic myosin. On a Sepharose CL 4B column ( $1.9 \times 82$  cm). The column was equilibrated and eluted with 0.5 M KCl containing 8 mM  $\text{KHCO}_3$ . 46 mg uc-myosin was applied to the column in 6 ml volume. Flow rate, 18–20 ml/h. The protein contents of tubes were detected via uv absorbance at 280 nm. The first peak (tubes 14–19, 20 mg) was pooled as myosin



Table 1

*Yields of chloroplast and myosin, and phosphate content of myosin prepared from spinach leaves*

Expt. No.	Mass of spinach leaves (wet weight) g	Yield of chloroplastic protein mg	Yield of gel-filtered myosin		Total P content of myosin mol P/mol	P content of lipid-free myosin mol P/mol	Lipid P content in myosin mol P/mol	Ribose P content in myosin mol P/mol
			mg	%				
1	1250	1526	14.56	0.955	39.6	33.96	3.8	Trace (0.8)
2	2000	2339	19.99	0.855	35.1	29.1	2.85	3.0
3	1000	1700	15.5	0.906	31.4	24.2	3.6	2.7
4	1100	2030	18.2	0.896	28.8	23.9	3.1	≈1

in the chloroplast may be about 2%. The last stage of purification was gel filtration, as illustrated by the elution profile of myosin in Figure 1.

It can be seen that  $\mu$ c-myosin contains a significant amount of low molecular mass protein, with traces of chlorophyll. The tubes of the first peak were collected as myosin (20 ccm) free from chlorophyll. The analytical data on four successive preparations are given in Table 1.

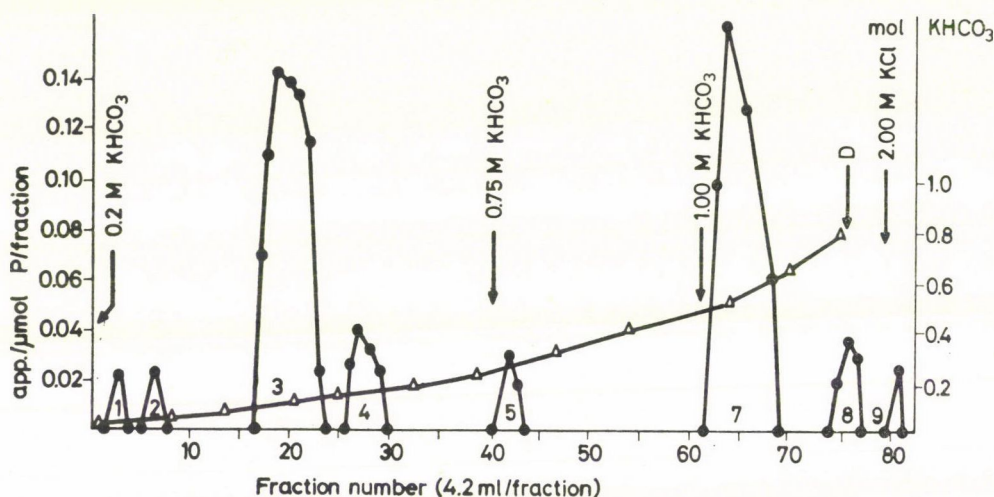


Fig. 2. Elution profile of P-containing fractions from gel-filtered myosin hydrolysate. 9.5 mg lipid-free protein was hydrolysed in a teflon ampoule in the presence of 3 M KOH (105 °C, 10 h). The hydrolysate was diluted to 50 ml and an aliquot used for P content determination before application to the column. The hydrolysate containing 0.792  $\mu$ mol P (apparently  $\approx$  3.17  $\mu$ mol P) was then diluted to 0.005 M KOH concentration and percolated through a Dowex 1X8 column (0.9  $\times$  6 cm). The separation was performed with a linear, but consecutive gradient chromatographic technique using a mixing chamber with a capacity of 160 cm<sup>3</sup> containing 0.02 M KHCO<sub>3</sub> and 120 ml of 0.2 M KHCO<sub>3</sub> in the reservoir equipped with an electromagnetic stirrer. The KHCO<sub>3</sub> concentration was increased and changed in reservoir as indicated by the arrows. 1 M KHCO<sub>3</sub> was applied to the column directly (D) without mixing, and 2 M KCl for regeneration of the column. The P content was determined in each effluent tube, in the presence of HClO<sub>4</sub>. The P contents of tubes are presented as  $\mu$ mol per fraction

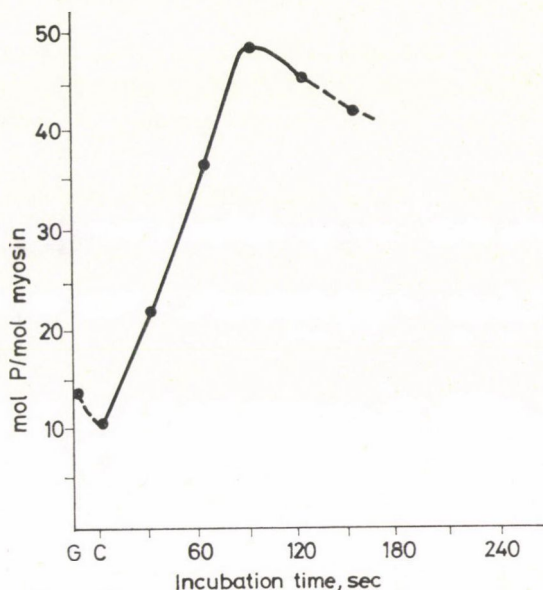


Fig. 3. Effect of incubation time on phosphate saturation of gel-filtered myosin. 0.48 mg gel-filtered myosin sample was incubated in each medium of 2.5 ml final volume. Composition of medium: 120 mM KCl, 25 mM Tris-HCl buffer, pH 7.28, 10 mM  $MgCl_2$ ,  $5 \times 10^{-5}$  M  $CaCl_2$ , 60 mM NaCl, 2.8 mM ATP. The incubation was started by addition of ATP, carried out at 30 °C, and terminated at the appropriate time with ice-cooled acetone. The samples were stored in a refrigerator overnight to precipitate all the protein. The protein was sedimented by centrifugation and excess nucleotide was removed with "washing solution", followed by the removal of lipids with  $CHCl_3$  - MeOH (2 : 1 v/v). The lipid-free samples were combusted in the presence of cc  $HNO_3$ , and the inorganic residues were applied for P determination. The P (including the incorporated P) contents of samples are presented as mol P/mol myosin versus incubation time. G ... gelfiltered myosin, C ... control, after preincubation without ATP

It was observed (Table 1) that the chloroplastic myosin contained a significant amount of endogenous phosphate. Therefore, the alkaline hydrolysates of lipid-free myosins were applied to a Dowex  $1 \times 8$  column and subjected to ion-exchange chromatography. As shown by Figure 2, the effluent fractions generally yielded 7-9 P-containing peaks, and they were numbered in order of elution.

Earlier evidences on Figure 2 are detailed in (Fazekas et al. 1981).

It can be seen in Figure 2 that P-Arg (3) and N-P-His (7) accounted for a considerable proportion of the P-containing peaks from freshly gel-filtered myosin.

Table 2 demonstrates the distribution and percentages of P-containing fractions, as revealed by the profile in Figure 2.

Our earlier studies showed that the binding sites of myosin were not completely saturated (Fazekas et al. 1982). Therefore, all myosin preparations



**Table 2**  
*Distribution of P-containing fractions in hydrolysate  
of lipid-free myosin*

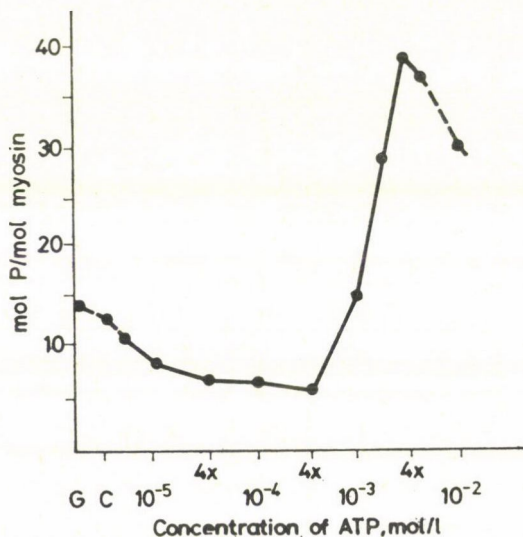
No.	Sample	Apparent $\mu\text{mol P}^+$	%
1	?	0.020	0.67
2	Phosphoramidate	0.032	1.08
3	P-Arg	1.300	43.88
4	Pi	0.065	2.19
5	P-Lys	0.040	1.35
6	?	—	Trace
7	N <sup><math>\alpha</math></sup> -P-His	1.420	47.94
8	N <sup><math>\epsilon</math></sup> -P-His	0.050	1.68
9	?	0.020	0.67
Total P		2.947	**

+ The actual P content was determined before chromatographic separation in the aliquot of the hydrolysate of 9.5 mg protein containing 0.792  $\mu\text{mol P}$  (corresponding to apparent 3.168  $\mu\text{mol P}$ ).

\*\* Recovery corresponds to about 93%.

were able to bind more phosphoryl groups by autophosphorylation from an ATP-containing suitable incubation medium.

The extent of phosphorylation depends on the incubation time (Figure 3).



*Fig. 4.* Phosphorylation of chloroplastic myosin in presence of increasing concentration of ATP. The incubation medium was the same as in Figure 3. The reaction was started by the addition of ATP and terminated at 90 s as an appropriate incubation time. Other legends as in Figure 3

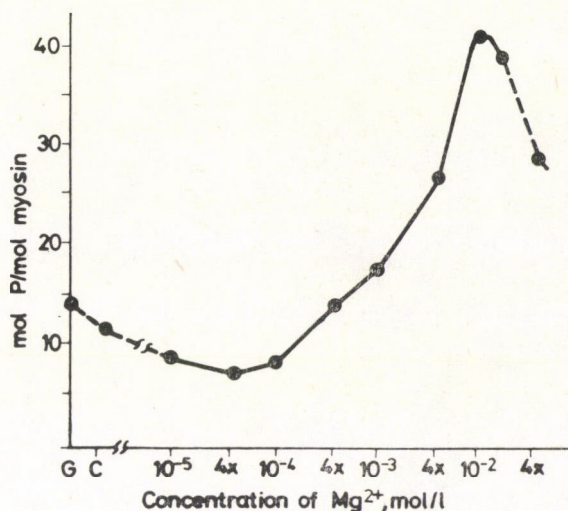


Fig. 5. Phosphate incorporation as a function of Mg ion concentration in the incubation medium. The reaction mixture was the same as in Figure 3, except that the Mg ion concentration was varied. Other conditions were the same as in Figure 3

Figure 3 shows that maximum saturation was reached with a short incubation, in about 90 s. The incorporated P may be stabilized by ice-cooled acetone precipitation and applied for P content determination.

The P saturation depended on the ATP concentration too, as shown by Figure 4.

During the preincubation and also in the presence of a low ATP concentration a declining P content was found while in a higher ATP concentration range the saturation increased reaching a maximum at about the physiological concentration (2–4 mM).

The phosphate saturation was  $Mg^{2+}$ -dependent. Figure 5 shows the myosin phosphate uptake as a function of the  $Mg^{2+}$  concentration. The maximum P incorporation was reached at a concentration of around 10 mM  $Mg^{2+}$ .

The curves of Figures 3–5 have minima and maxima. The minimum indicates the effects of storage and incubation. This myosin already differed slightly from the gel-filtered one. The organic P content decreased with aging. The energy rich phosphates were more labile (e.g. P-Arg). The declined parts of curves indicate the end product inhibition (Fazekas et al. 1981).

The chloroplastic myosin was able to form a characteristic filamentous aggregating system. At the beginning of filament formation induced by 1 mM  $K[Au(CN)_4]$ , the filaments were so thin that the arrangement of the individual molecules could be observed, as in Figure 6.





*Fig. 6.* Formation of filamentous aggregate system from gel-filtered myosin, induced by 1 mM  $K[Au/CN_4]$  solution. The preparation was postcontrasted by  $AuCl_3$  treatment. Final magnification, X 18.000 (Photo: Óváry)

### Discussion

These experiments demonstrate that 0.8–1% of the chloroplast proteins can be recovered as myosin, if the loss involved in the myosin comprises 1.6–2% of the total protein in the chloroplast.

The freshly gel-filtered and lipid-free myosin preparation contains an appreciable amount of endogenous phosphate. This is mainly in the form of N-P-bonded phosphoryl groups with a high transfer potential. By means of ion-exchange chromatography, the phosphate in an alkaline hydrolysate of the preparation can be separated into 7–9 P-containing fractions. In this, it conforms with the previously examined myosin prepared from various sources, but chloroplast myosin differs in that the  $N^{\gamma}$ -P-His (7) peak (fraction is the largest, and also for P-Arg. During the identification, fraction 2 proved



to be phosphoramidate, which is the final hydrolytic product of P-Arg. Its quantity is definitely dependent on the duration of protein hydrolysis. The origin of fractions 1 and 6 are uncertain. Peak 9 does not relate to one definite P-containing compound, for it is obtained by regeneration of the ion-exchange column. Relatively insignificant minor peaks (comprising only a few per cent) (peaks 2a and 7a) appear if a larger amount of P-containing hydrolysate (1.5–3  $\mu\text{mol}$ ) is separated on the chromatographic column. Their origins are uncertain, since the course of the alkaline hydrolysis on the proteins has not yet been clarified.

The P content of the freshly gel-filtered myosin preparations can be increased by phosphorylation. The degree of phosphorylation depends on the ATP and  $\text{Mg}^{2+}$  concentrations of the medium, and also on the duration of the incubation.

Myosin preparations that have stood for some time incorporate either no phosphoryl groups or scarcely any. However, their ATPase activity (hydrolytic enzyme fraction) remains. These filamentary systems, such as shown in Figure 6, cannot be formed from such preparations, but only coarse aggregates.

We believe that, besides the chloroplast myosin, cytoplasmic myosin is also present in the cells of leaves. This has not yet been separated, and thus its concentration is unknown.

### Acknowledgement

The authors are very grateful to Professor Antoni for his deep interest and valuable advice, and to Mrs. Bökönyi for skilful technical assistance.

### References

- Briat, J. F., Lesquire, A. M., Mache, R. (1986): Transcription of chloroplast DNA. A review. *Biochem. (Paris)*, **68**, 981–990.
- Fazekas, S., Fehér, J., Kondics, L., Székessy-Hermann, V. (1987): Preparation and characterization of mitochondrial myosins of rat and human liver. *Acta Physiol. Hung.*, **70**, 3–24.
- Fazekas, S., Óváry, I., Horváth, E., Székessy-Hermann V., Juhász, P. (1982): Isolation, properties and P content of the human brain myosin. *Acta Physiol. Hung.*, **59**, 101–117.
- Fazekas, S., Samu, J., Szabó, E., Székessy-Hermann, V. (1981): Identification and specific reaction of alkali stable amino acid phosphates in myosin hydrolysates. *Acta Agron. Hung.*, **30**, 340–350.
- Kroon, A. M. (1969): *DNA and RNA from mitochondria and mitoplasts*. In Lima-de Faizired, A. (ed): Handbook of Molecular Cytology. North-Holland Publisher, Amsterdam. 943–971.
- Menzel, D., Schliwa, M. (1986): Motility in the *Siphonous* green alga *Bryopsis*. II. Chloroplast movement requires organized arrays of both microfibrilles and actin filaments. *Eur. J. Biol.*, **40**, 286–295.
- Nehéz, R., Fazekas, S., Óváry, I., Székessy-Hermann, V. (1980): *Purification and some properties of myosin prepared from root tips of Zea mays L.* (SZE DC 384 hybrid). Proc. 20th Ann. Meet. Biochem. HCS. Siófok. 245–245.
- Nehéz, R., Fazekas, S., Óváry, I., Székessy-Hermann, V. (1985): Purification and some properties of myosin prepared from root tips of maize (*Zea mays L.*) seedlings. *Acta Agrom. Hung.*, **34**, 267–273.



- Nehéz, R., Fazekas, S., Óváry, I., Székessy-Hermann, V. (1986): Myosin preparations from the meristematic tissue of fresh sprouts of vine (*Vitis vinifera* L., cv. Cardinal). *Acta Agron. Hung.*, **35**, 3-10.
- Onishi, T. (1964): Le changement de volume du chloroplaste, accompagné de phosphorylation, et les protéines ressemblantes à l'actine et à la myosine extraites du chloroplaste. *J. Biochem. (Tokyo)*, **55**, 494-505.
- Schnaitman, G., Greenwalt, J. W. (1968): Enzymatic properties of the inner and outer membranes of rat liver mitochondria. *J. Cell. Biol.* **38**, 158-175.
- Schorer-Mörtel, G. (1972): Reversible Form und Volumenänderungen des *Mougeotia*-Chloroplasten. II. Dunkelheit sowie Beziehungen zu den Orientierungsbewegungen. *Z. Pflanzenphysiol.*, **68**, 93-214.
- Schönboom, E. (1973): Kontraktile Fibrillen als aktive Filament beider Mechanik der Chloroplasten-Verlieferung. *Br. Dtsch. Bot. Ges.*, **84**, 407-422.
- Sokolov, O., I., Bogatyrev, V. A., Turkina, M. V. (1986): Myosin from conducting tissues of *Heracleum sosnowskyi*: Interaction with muscle actin and formation of filaments. (In Russian.) *Fiziologija rashtenij*. **33**, 421-431.
- Ster, D. F., Palmer, J. D. (1984): Extensive and widespread homologies between mitochondrial DNA and chloroplast DNA in plants. *Proc. Natl. Acad. Sci. US.*, **81**, 1946-1950.
- Tandler, B., Hoppel, Ch. L. (1972): *Mitochondria*. Monography. Academic Press, New York and London.
- Wagner, G., Haupt, W., Laux, A. (1972): Reversible inhibition on chloroplast movement by cytochalasin B in the green alga *Mougeotia*. *Science*, **176**, 808-809.
- Walker, D. A. (1980): Preparation of higher plant chloroplasts. *Methods in Enzymol.*, **69**, 94-104.
- Williamson, P. E. (1976): *Actin and motility in plant cells*. In Perry, S. V., Margreth, A., Adelstein, R. S. (eds): *Contractile systems in non muscle tissues*. Elsevier, Amsterdam, Oxford, New York, 91-101.

## EFFECT OF FERTILIZATION ON NUTRIENT UPTAKE AND DISTRIBUTION IN WINTER WHEAT DURING VEGETATION

B. LÁSZTITY

RESEARCH INSTITUTE FOR SOIL SCIENCE AND AGROCHEMISTRY OF THE  
HUNGARIAN ACADEMY OF SCIENCES, BUDAPEST, HUNGARY

(Received 14 October 1987; accepted 4 January 1988)

In a field fertilization experiment carried out on calcareous chernozem soil, the uptake of N, P, K, Ca, and Mg in the winter wheat leaf, stalk, ear, grain and in the straw was studied during vegetation period. The samples were taken from 0.5 m<sup>2</sup> of plot, generally every ten days from spring until harvesting. The results can be summarized as follows.

In the leaves the uptake of N, P, K and Ca reached maximum in the period of the vegetative growth and of the Mg in the flowering stage. In the stalk the uptake of N, P and Ca reached maximum at the flowering, while of K and Mg in the milky ripening phase. In the ear all the elements reached maximum in milky ripening phenophase.

The NPK fertilization increased the uptake of N, P, K, Ca and Mg compared to the control in all examined plant parts.

**Keywords:** NPK fertilization, N, P, K, Ca, Mg uptake and distribution, vegetation period, winter wheat

### Introduction

The intensity of uptake of the individual nutritive elements — and their accumulation in different plant parts during the vegetation period — generally is a somewhat neglected research field of agrochemistry and plant nutrition. The quantitative relations of accumulation supply useful information for the planning of plant nutrition and the proper use of fertilizers. At the same time they fill a gap existing in this respect in the relevant literature.

The quantitative relations of material turnover, and of translocation between plant parts within, are adjusted to the biological functions and are characteristic of the species, variety and variety group concerned (Pethő 1984, Sarič 1981). They also indicate the direction of the accumulation processes and point to the nutrient status of the plant (Biczók and Lásztitý 1985, Potapov and Dézsi 1954).

The papers written about the distribution of nutrient accumulation within the plant discuss the subject primarily in relation to the phenophase of full maturity (Ellen 1987, Kádár et al. 1985, Sarkadi et al. 1968). A relatively large number of publications examining the process of fruit forming deal with nutrient accumulation in various plant parts — particularly in the grain — in the period following flowering (Adorján 1902, Kiss et al. 1984).



Most of the data concerning the distribution of nutrients refer to macroelements and only an insignificant part of them to microelements.

As to the relationship between nutrient distribution in plants and fertilization, information is mostly available on the accumulation of nitrogen. Data on the other macroelements, and on microelements, are substantially smaller in number (Biczók et al. 1985, Gregory et al. 1979, Láng 1960). Concerning changes of accumulation in the different plant parts during vegetation little is known. For this reason we propose to supply data on the dynamic of nutrient uptake and distribution in winter wheat in the case of a varying rate and composition of fertilization.

### Materials and methods

The experiments were carried out under field conditions. The fertilization experiment was set up on calcareous chernozem soil at the Experiment Station of the Hungarian Academy of Sciences, Nagyhorcsók. The plant material to be examined — the total aboveground part — was taken from 4 running metres per plot on 10 occasions from spring to harvesting.

On the first two occasions that the entire plant was weighed, we found no reason for separating the plant parts. From the third occasion of sampling the stalks and leaves, from the sixth sampling the ears too, and at the full ripening the grains and the vegetative parts were separated and weighed in two replications. The plant material weighed was analysed after due preparation. Using the dry matter production and analysis data the quantity taken up was calculated for each plant part and for the entire aboveground part of plant. Data are expressed by elements and on absolute dry matter basis.

In the fertilization treatments combinations of 200 kg nitrogen (N), 500 kg  $P_2O_5$  ( $P_1$ ) and  $K_2O$  ( $K_1$ ) respectively, and 1000 kg  $P_2O_5$  ( $P_2$ ) and  $K_2O$  ( $K_2$ ) per ha quantities of fertilizer were used. The doses were chosen with the view of ensuring different levels of P- and K-supply within the experiment. As fertilizer *pétisó* (28% N) superphosphate (17%  $P_2O_5$ ) and potassium salt (40%  $K_2O$ ) were used.

The winter wheat variety was "Martonvásári 8". The biometric evaluation was carried out by variance analysis. On the other conditions of the experiment information has already been supplied in a previous publication (Lásztity 1987).

### Results

In order to trace the nutrient uptake by plant parts in Fig. 1 we show the trend of dry matter production during vegetation in the major plant parts.

The nitrogen uptake by the individual plant parts of winter wheat in the different treatment is shown in Table 1, and the averages are seen in Fig. 2. In the leaf the uptake reaches its maximum — 63 kg/ha — at the end of shooting, then falls to 14 kg/ha by the time of full maturity. The share of uptake within the entire aboveground plant part increases from the initial 54% to 67%, then falls back to 10% before harvesting. Fertilization significantly increased the uptake compared to the control, in the NP- and NPK

treatments consistently and in the N- and NK treatments with a single exception. PK treatment without nitrogen did not ensure any increase in uptake.

In the stalk the nitrogen uptake steadily increased and reached its maximum in the phenophase of earing, then decreased until maturing. Its relative value within the total aboveground part ranged between 46% and 23%, with comparatively wide fluctuations in the course of vegetation. On each occasion of sampling, fertilization was found to have resulted in a significant increase in uptake compared to the control. A significant increase was found in the NPK treatment at each time of sampling, in the NP treatment with one and in the N treatment with two exceptions: in the NK treatment significant increase was only found on a few occasions while the PK treatment

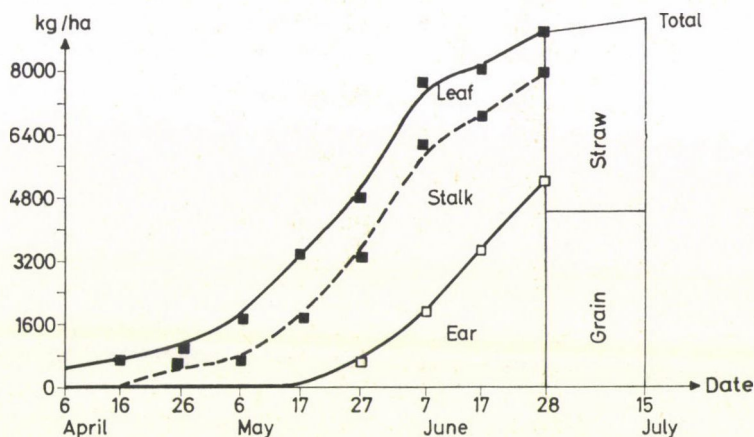


Fig. 1. Dry matter accumulation in winter wheat 1982

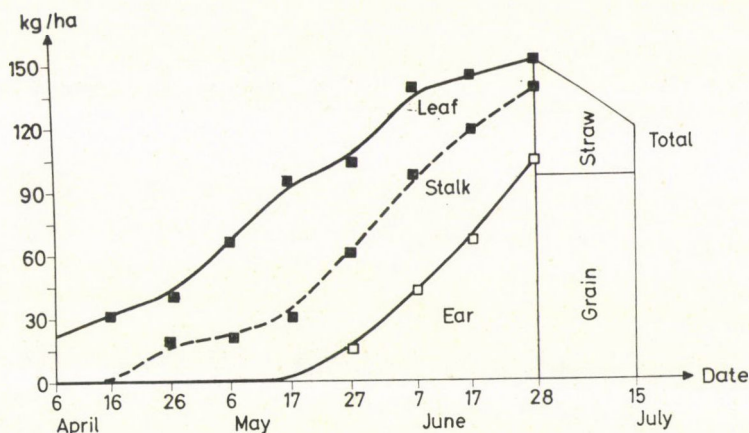


Fig. 2. N-uptake by winter wheat 1982



Table 1

*Effect of fertilization on nitrogen uptake by winter wheat (kg/ha) in various phases of growth*

*Variety: "Martonvásári 8." Nagyhörcsök 1982*

Treatment	Time of sampling				Development phase (feekes)		
	April 26 (6)	May			June		
		6 (7)	17 (8)	27 (10)	7 (10.1)	17 (10.5)	28 (11.1)
Leaf							
1. Ø	10.5	19.8	28.3	24.1	15.5	12.6	5.9
2. N	18.2	37.5	68.1	54.6	51.1	27.8	16.4
3. P <sub>1</sub> K <sub>1</sub>	14.6	24.2	23.4	18.8	17.3	9.9	6.7
4. NP <sub>1</sub>	30.2	64.6	87.3	61.4	50.6	33.3	19.1
5. NK <sub>1</sub>	14.2	40.4	63.2	47.9	48.8	30.6	13.8
6. NP <sub>1</sub> K <sub>1</sub>	38.6	60.5	89.3	49.8	57.0	31.7	18.6
7. NP <sub>2</sub> K <sub>2</sub>	30.8	63.8	79.9	47.8	40.4	31.0	13.9
LSD <sub>5%</sub>	8.9	16.2	15.8	15.1	12.4	7.3	4.4
Average	22.4	44.4	62.8	43.5	40.1	25.3	13.5
% of total uptake	54	67	67	42	29	18	9
Stalk							
1. Ø	8.1	7.4	20.3	19.5	33.3	33.5	38.5
2. N	10.2	28.8	40.2	51.7	55.9	66.4	44.1
3. P <sub>1</sub> K <sub>1</sub>	9.3	10.7	19.9	24.8	20.0	35.4	14.3
4. NP <sub>1</sub>	35.6	33.9	35.9	51.3	56.2	60.6	46.6
5. NK <sub>1</sub>	11.7	14.9	27.3	47.1	89.7	43.2	19.3
6. NP <sub>1</sub> K <sub>1</sub>	30.8	35.2	42.9	64.9	49.9	54.7	29.0
7. NP <sub>2</sub> K <sub>2</sub>	25.8	23.0	29.8	58.9	76.0	69.0	49.1
LSD <sub>5%</sub>	9.1	10.5	9.1	13.6	13.0	13.7	9.5
Average	18.8	22.0	30.9	45.4	54.4	51.8	34.4
% of total uptake	46	33	33	44	40	36	23
Ear							
1. Ø	—	—	—	10.3	29.7	41.3	63.1
2. N	—	—	—	12.5	42.1	64.8	103.8
3. P <sub>1</sub> K <sub>1</sub>	—	—	—	9.5	29.3	44.7	75.7
4. NP <sub>1</sub>	—	—	—	20.8	49.8	71.0	121.4
5. NK <sub>1</sub>	—	—	—	12.4	45.8	71.9	112.6
6. NP <sub>1</sub> K <sub>1</sub>	—	—	—	20.1	45.3	94.5	127.7
7. NP <sub>2</sub> K <sub>2</sub>	—	—	—	21.4	58.2	81.0	121.8
LSD <sub>5%</sub>	—	—	—	4.1	10.8	19.2	27.9
Average	—	—	—	15.3	42.9	67.0	103.7
% of total uptake	—	—	—	14	31	46	68

caused depression. In the ear the quantity of nitrogen taken up gradually increased with the advance of vegetation from 15 to 103 kg/ha, and its relative value within the plant grew from 14% to 68% by the end of milky ripeness. Fertilization resulted in a significant increase of uptake, as found on each sampling in the NP and NPK treatments and with one exception in the N and NK treatments. The PK treatment proved ineffective. On harvesting a large proportion (Fig. 2), more than 80% of the nitrogen taken up was stored in the grains and only a minor part was located in the vegetative parts.

The trend of phosphorus uptake by the winter wheat in the course of vegetation is shown in Table 2, and on the average of the experiment in Fig. 3.

In the total aboveground part and in the leaf an increase in phosphorus uptake can be observed with the advance of vegetation, which in the leaf reaches maximum at the end of shooting. After that it tends to decrease both as regards the absolute quantity and the value relative to the aboveground part.

Fertilization was found to increase the uptake on each occasion of sampling in the NP- and NPK treatments, and with one and two exceptions in the N and NK treatments, respectively. The PK treatment only increased the uptake compared to the control in the initial phases of growth.

In the stalk the phosphorus uptake during the vegetation period ranged between 1.5 and 8.0 kg/ha. After the highest rate of uptake in the phenophase of flowering, a decreasing tendency was observed. Owing to the translocation during vegetation its relative value fell from 66% to 8%. As to the effect of fertilization, the influence of NP succeeded on each sampling, the influence of nitrogen was demonstrable in the majority of the samples, while the effect of NK only could be pointed out in several cases.

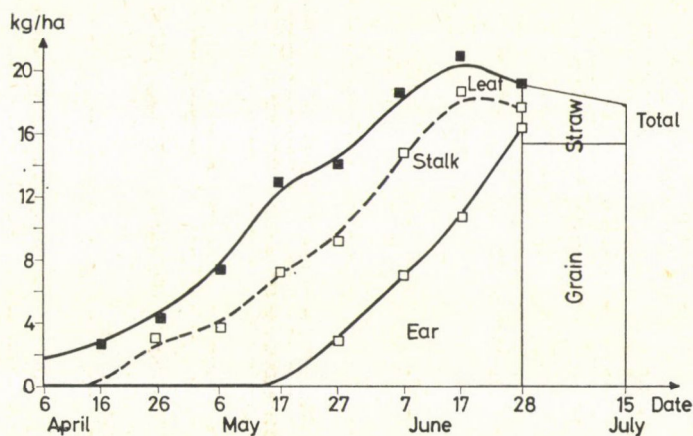


Fig. 3. P-uptake by winter wheat 1982



Table 2

*Effect of fertilization on phosphorus uptake by winter wheat (kg/ha)  
in various phases of development  
Variety: "Martonvásári 8" Nagyhöröcsök, 1982*

Treatment	Time of sampling				Development phase (feekes)		
	April 26 (6)	May			June		
		6 (7)	17 (8)	27 (10)	7 (10.1)	17 (10.5)	28 (11.1)
<i>Leaf</i>							
1. Ø	0.7	1.8	3.1	2.9	2.0	1.5	0.9
2. N	1.3	2.8	5.9	5.5	4.3	2.7	1.8
3. P <sub>1</sub> K <sub>1</sub>	1.4	2.4	2.8	2.7	1.9	1.1	1.0
4. NP <sub>1</sub>	2.0	5.3	7.6	6.2	5.0	2.7	2.0
5. NK <sub>1</sub>	0.9	3.4	5.2	5.1	4.2	2.3	1.5
6. NP <sub>1</sub> K <sub>1</sub>	2.7	4.7	8.4	5.6	5.4	2.9	2.0
7. NP <sub>2</sub> K <sub>2</sub>	2.0	4.8	6.6	5.2	4.2	2.4	1.5
LSD 5%	0.6	1.3	1.4	1.3	1.0	0.5	0.5
Average	1.5	3.6	5.7	4.9	3.7	2.2	1.5
% of total uptake	34	49	45	35	20	11	8
<i>Stalk</i>							
1. Ø	1.4	1.6	4.7	3.9	5.3	6.9	0.8
2. N	2.2	3.7	5.8	7.3	6.7	8.8	1.0
3. P <sub>1</sub> K <sub>1</sub>	2.0	2.2	5.9	4.9	5.9	6.7	0.1
4. NP <sub>1</sub>	5.0	6.1	9.5	6.1	8.3	9.3	2.0
5. NK <sub>1</sub>	1.4	2.1	6.4	6.3	9.3	6.5	0.8
6. NP <sub>1</sub> K <sub>1</sub>	4.2	5.8	9.0	8.1	7.4	8.1	2.2
7. NP <sub>2</sub> K <sub>2</sub>	3.9	4.9	8.4	8.1	10.1	9.0	2.6
LSD 5%	1.4	1.8	2.0	1.9	1.7	2.5	0.5
Average	2.9	3.8	7.1	6.3	7.7	7.9	1.3
% of total uptake	66	51	55	45	42	38	7
<i>Ear</i>							
1. Ø	—	—	—	2.0	4.8	7.5	11.1
2. N	—	—	—	2.3	6.6	9.6	15.9
3. P <sub>1</sub> K <sub>1</sub>	—	—	—	1.9	5.4	9.6	15.5
4. NP <sub>1</sub>	—	—	—	4.0	7.6	11.7	17.5
5. NK <sub>1</sub>	—	—	—	2.3	7.2	9.3	15.9
6. NP <sub>1</sub> K <sub>1</sub>	—	—	—	4.4	7.8	13.4	19.2
7. NP <sub>2</sub> K <sub>2</sub>	—	—	—	4.3	9.1	13.8	18.7
LSD 5%	—	—	—	0.8	1.6	2.0	4.5
Average	—	—	—	2.9	7.0	10.7	16.3
% of total uptake	—	—	—	20	38	51	85

Table 3

*Effect of fertilization on potassium uptake by winter wheat (kg/ha)  
in various phases of development  
Variety "Martonvásári 8" Nagyhőrcsök, 1982*

Treatment	Time of sampling				Development phase (feekes)		
	April 26 (6)	May			June		
		6 (7)	17 (8)	27 (10)	7 (10.1)	17 (10.5)	28 (11.1)
Leaf							
1. Ø	7.5	14.4	24.1	27.4	16.7	10.4	4.8
2. N	15.5	31.3	54.1	61.0	39.7	18.9	11.1
3. P <sub>1</sub> K <sub>1</sub>	12.7	27.4	23.4	25.7	18.1	9.9	5.7
4. NP <sub>1</sub>	23.8	51.5	67.6	68.0	46.2	21.5	12.8
5. NK <sub>1</sub>	10.2	32.2	54.8	58.4	45.9	16.3	11.3
6. NP <sub>1</sub> K <sub>1</sub>	30.0	45.0	71.0	60.6	51.8	19.7	13.5
7. NP <sub>2</sub> K <sub>2</sub>	21.6	42.6	60.2	59.7	37.0	20.9	11.9
LSD <sub>5%</sub>	7.1	12.3	15.5	18.0	11.3	4.9	4.6
Average	17.3	34.9	50.7	51.5	36.5	16.9	10.2
% of total uptake	49	60	54	39	27	14	14
Stalk							
1. Ø	6.9	9.8	26.6	36.5	50.1	61.7	30.6
2. N	12.3	20.1	22.9	53.7	63.5	73.2	36.4
3. P <sub>1</sub> K <sub>1</sub>	9.4	3.6	30.5	45.4	52.9	54.8	22.2
4. NP <sub>1</sub>	27.9	30.8	61.5	65.4	79.3	77.2	28.0
5. NK <sub>1</sub>	10.0	17.2	31.7	56.5	101.5	81.0	38.5
6. NP <sub>1</sub> K <sub>1</sub>	27.6	39.4	66.8	112.1	75.5	95.7	36.0
7. NP <sub>2</sub> K <sub>2</sub>	30.8	42.1	55.5	104.0	112.8	110.2	38.4
LSD <sub>5%</sub>	12.3	11.2	13.0	21.4	25.4	31.0	11.1
Average	17.9	23.3	42.3	67.6	76.5	79.4	32.8
% of total uptake	51	40	46	52	58	64	44
Ear							
1. Ø	—	—	—	7.3	13.2	19.0	21.4
2. N	—	—	—	9.2	20.6	27.6	33.9
3. P <sub>1</sub> K <sub>1</sub>	—	—	—	6.8	14.0	20.3	28.3
4. NP <sub>1</sub>	—	—	—	16.2	22.2	29.6	30.6
5. NK <sub>1</sub>	—	—	—	9.2	20.5	27.6	30.7
6. NP <sub>1</sub> K <sub>1</sub>	—	—	—	17.1	22.7	35.1	39.7
7. NP <sub>2</sub> K <sub>2</sub>	—	—	—	17.5	26.4	35.9	33.8
LSD <sub>5%</sub>	—	—	—	3.7	4.4	5.8	7.9
Average	—	—	—	11.9	19.9	28.1	31.2
% of total uptake	--	—	—	9	15	22	42



The phosphorus uptake by the ear steadily increased, on the average of the experiment from 3 to 16 kg/ha. The share of the accumulated phosphorus within the total aboveground part grew from 20% to 85%. Out of the fertilization treatments, NP and NPK proved to affect the phosphorus uptake favourably in all cases, N did so with one exception, while NK was found to be effective in half of the cases.

On full ripening the largest quantity of phosphorus was found in the grain, and only a quarter of the total amount accumulated in the vegetative parts (Fig. 3). Potassium accumulation in the different organs of winter wheat is shown in Table 3 while Fig. 4 illustrates the experiment average.

In the total aboveground part as well as in the leaf, the potassium accumulation increased parallel with the time up to the phase of earing. Afterwards a decrease could be observed with a minimum value of 10 kg/ha prior to harvesting. Its share within the total aboveground plant part fell from 60% to 14% between shooting and maturing, due partly to a loss and partly to the translocation. Fertilization was found on each occasion of sampling to have caused a significant increase in potassium uptake in the nitrogen (N, NP, NK, NPK) treatments. The PK treatment proved to result in a significant increase, compared to the control, only in a single case of sampling.

In the stalk the potassium accumulation increased until the end of earing, then decreased. The maximum 80 kg/ha — was measured at the end of earing. The relative value of potassium accumulation ranged between 40% and 64%, indicating that the largest quantity was accumulated in the stalk. Out of the fertilizer treatment, the effect of NP and NPK was significant though not at each time of sampling.

In the ear the potassium uptake increased in a positive correlation with the time and the maximum was measured before full ripening. Its relative

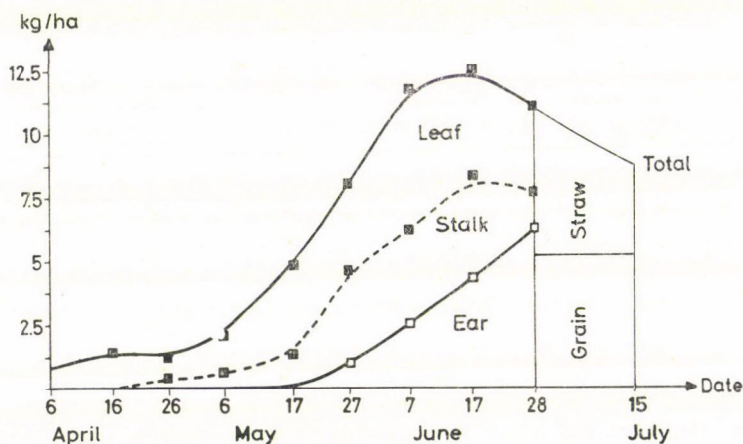


Fig. 4. K-uptake by winter wheat 1982

Table 4

*Effect of fertilization on calcium uptake by winter wheat (kg/ha)  
in various phases of development  
Variety "Martonvásári 8" Nagyhőrcsök 1982*

Treatment	Time of sampling				Development phase (feekes)		
	April	May			June		
	26 (6)	6 (7)	17 (8)	27 (10)	7 (10.1)	17 (10.5)	28 (11.1)
<i>Leaf</i>							
1. Ø	2.0	5.9	9.6	9.5	7.3	5.3	4.9
2. N	4.6	9.5	20.5	20.7	20.7	13.5	11.6
3. P <sub>1</sub> K <sub>1</sub>	5.0	8.0	8.6	9.0	8.2	5.7	4.2
4. NP <sub>1</sub>	7.6	15.7	22.7	25.0	20.6	14.9	12.1
5. NK <sub>1</sub>	3.3	8.7	14.2	18.4	17.5	12.6	10.2
6. NP <sub>1</sub> K <sub>1</sub>	9.1	15.4	24.4	27.3	22.9	14.9	10.7
7. NP <sub>2</sub> K <sub>2</sub>	7.3	15.5	22.7	20.2	20.5	13.3	10.5
LSD <sub>5%</sub>	2.2	4.1	5.5	5.3	5.3	3.0	2.7
Average	5.6	11.3	17.9	18.6	16.8	11.5	9.2
% of total uptake	82	92	92	93	66	58	63
<i>Stalk</i>							
1. Ø	1.4	0.6	0.9	1.0	7.5	9.4	4.7
2. N	2.0	2.6	0.7	0.2	3.6	2.1	5.7
3. P <sub>1</sub> K <sub>1</sub>	0.1	0.2	0.9	1.9	4.8	7.5	3.6
4. NP <sub>1</sub>	2.3	1.8	4.5	0.1	8.4	6.2	0.2
5. NK <sub>1</sub>	0.4	0.1	0.5	0.6	12.4	2.2	2.3
6. NP <sub>1</sub> K <sub>1</sub>	1.7	1.4	3.5	0.1	4.8	8.1	3.2
7. NP <sub>2</sub> K <sub>2</sub>	1.0	0.8	0.1	0.2	5.0	8.3	2.8
LSD <sub>5%</sub>	0.7	0.5	0.8	0.2	2.1	1.5	0.8
Average	1.2	1.0	1.5	0.6	6.6	6.2	3.2
% of total uptake	18	8	8	3	26	31	22
<i>Ear</i>							
1. Ø	—	—	—	0.7	1.5	1.0	1.5
2. N	—	—	—	0.5	2.7	2.0	2.1
3. P <sub>1</sub> K <sub>1</sub>	—	—	—	0.7	1.4	1.2	2.7
4. NP <sub>1</sub>	—	—	—	0.7	2.1	2.6	2.1
5. NK <sub>1</sub>	—	—	—	0.6	1.9	1.9	2.1
6. NP <sub>1</sub> K <sub>1</sub>	—	—	—	1.1	2.6	3.3	3.1
7. NP <sub>2</sub> K <sub>2</sub>	—	—	—	1.1	2.5	2.3	1.8
LSD <sub>5%</sub>	—	—	—	0.3	0.6	0.5	1.2
Average	—	—	—	0.7	2.1	2.0	2.2
% of total uptake	—	—	—	4	8	11	15



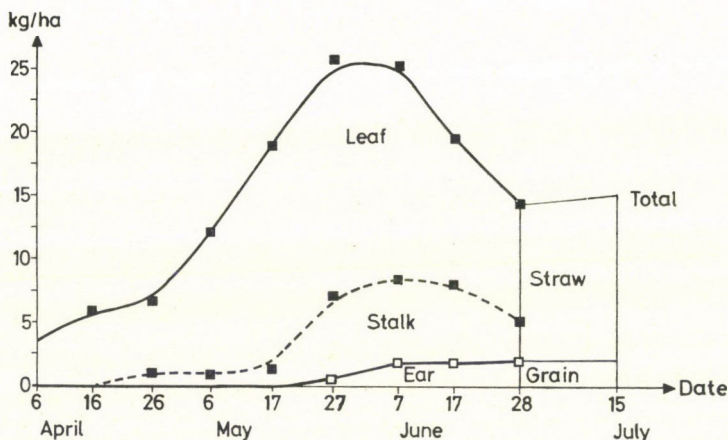


Fig. 5. Ca-uptake by winter wheat 1982

share within the total aboveground plant part grew from 9% to 42%. Fertilization showed a demonstrable increase in the nitrogen treatments in all cases of sampling. On harvesting the potassium accumulation in the vegetative parts considerably exceeded the quantity found in the grain yield (Fig. 4).

The calcium uptake by and its distribution in winter wheat are shown in Table 4, while Fig. 5 illustrates the experiment averages. In the early phase of development, the accumulation of calcium increased in the total aboveground part of plant, as it also did in the leaf until the end of earing. The maximum uptake was nearly 20 kg/ha. Having reached its maximum the calcium uptake showed the opposite tendency, and by the time of maturity a loss of this element was registered. It was in the leaf of all aboveground organs that the wheat plant accumulated the larger part of calcium at the time of maximum accumulation 90% of the total amount. Fertilization was found to cause an increase in calcium uptake in the NP-, NPK- and N treatments in all cases of sampling, while in the NK treatment only in the majority of the sampling occasions.

In the stalk the maximum and minimum values of calcium uptake differ about tenfold. The relative value of accumulation in the plant ranged between 3% and 31%. The highest value of accumulation was measured after flowering and the lowest on earing. The effect of fertilization was less consistent, since a significant increase in calcium uptake in the fertilization treatments was only observed in less than half of the sampling cases.

In the ear the accumulation of calcium increased with the advance of the vegetation period, from 0.7 to 2.2 kg/ha on the average of the experiment. Its relative value within the entire plant rose from 4% to 15%. The effect of fertilization was statistically proved in the NPK treatment on all occasions of sampling, in the N-, NP- and NK treatments in half of the sampling cases.

Table 5

*Effect of fertilization on magnesium uptake by winter wheat (kg/ha)  
in various phases of development  
Variety: "Martonvásári 8" Nagyhorcsók, 1982*

Treatment	Time of sampling				Development phase (feekes)		
	April	May			June		
	26 (6)	6 (7)	17 (8)	27 (10)	7 (10.1)	17 (10.5)	28 (11.1)
<i>Leaf</i>							
1. Ø	0.3	0.7	2.2	1.6	2.4	2.0	1.6
2. N	0.7	1.1	3.6	4.1	6.6	4.9	3.9
3. P <sub>1</sub> K <sub>1</sub>	0.6	1.0	1.5	1.5	2.3	1.6	1.3
4. NP <sub>1</sub>	1.2	2.4	5.1	4.7	6.9	5.4	4.2
5. NK <sub>1</sub>	0.5	1.2	3.1	3.6	6.3	5.0	3.7
6. NP <sub>1</sub> K <sub>1</sub>	1.3	2.1	4.9	5.0	7.9	5.4	4.4
7. NP <sub>2</sub> K <sub>2</sub>	1.1	2.2	4.1	3.6	6.2	5.4	4.2
LSD <sub>5%</sub>	0.3	0.8	1.1	1.0	1.4	1.3	1.1
Average	0.8	1.5	3.5	3.4	5.5	4.0	3.3
% of total uptake	67	71	73	42	47	34	30
<i>Stalk</i>							
1. Ø	0.3	0.2	0.2	1.8	1.5	3.0	1.3
2. N	0.4	1.0	1.1	3.8	2.8	5.4	1.0
3. P <sub>1</sub> K <sub>1</sub>	0.2	0.2	1.3	2.2	2.7	2.8	0.8
4. NP <sub>1</sub>	0.7	1.0	3.1	4.9	4.5	4.2	0.2
5. NK <sub>1</sub>	0.2	0.5	0.9	3.3	5.3	1.7	0.7
6. NP <sub>1</sub> K <sub>1</sub>	0.6	0.6	0.9	4.7	3.6	3.7	5.1
7. NP <sub>2</sub> K <sub>2</sub>	0.4	0.8	1.3	4.2	5.2	7.0	0.5
LSD <sub>5%</sub>	0.2	0.3	0.4	1.1	1.2	1.6	0.5
Average	0.4	0.6	1.3	3.6	3.7	4.0	1.4
% of total uptake	33	29	27	45	32	32	13
<i>Ear</i>							
1. Ø	—	—	—	0.7	1.8	2.9	4.1
2. N	—	—	—	0.8	2.9	3.9	6.7
3. P <sub>1</sub> K <sub>1</sub>	—	—	—	0.6	1.8	3.5	5.8
4. NP <sub>1</sub>	—	—	—	1.2	2.3	4.4	6.6
5. NK <sub>1</sub>	—	—	—	0.6	2.2	5.0	4.9
6. NP <sub>1</sub> K <sub>1</sub>	—	—	—	1.3	2.8	5.0	8.1
7. NP <sub>2</sub> K <sub>2</sub>	—	—	—	1.6	3.6	5.5	7.8
LSD <sub>5%</sub>	—	—	—	0.3	0.7	1.3	1.8
Average	—	—	—	1.0	2.5	4.3	6.3
% of total uptake	—	—	—	13	21	34	57



At the time of harvesting a decisive amount of Ca present in the above-ground part of plant was located in the vegetative parts (Fig. 5).

The magnesium uptake and distribution in the winter wheat examined is shown in Table 5 for each treatment, and in Fig. 6 for the average of the treatments.

In the aboveground part of the young plant as well as in the leaf, the magnesium accumulation increased up to the phenophase of flowering. Following the maximum of accumulation until maturity, a gradual decrease could be observed. The share of leaves from magnesium accumulation compared to the total aboveground part ranged between 30% and 73%, indicating that in the phase of vegetative growth the larger part of the amount of magnesium which was taken up had accumulated in the leaves. The influence of fertilization could be statistically proved; it was significant on all occasions of sampling in the NP- and NPK treatments, with one exception in the N treatment, and in less than half of the case of sampling in the N treatment. In the stalk the accumulation of Mg showed a similar tendency to that of the leaf, the difference being only found in the absolute values. Its relative value in the total aboveground part ranged between 45% and 13% in the course of vegetation, indicating that only a smaller proportion of the accumulated magnesium is located in the stalk.

It was only in the NPK treatment that the favourable effect of fertilization could be observed at each time of sampling. In the other (NP, NK, N) treatments the effect of fertilization was not consistently significant.

In the ear the accumulation of magnesium proved to be continuous. The amount of magnesium taken up grew from 1 to 6 kg/ha on the average of the experiment. During the same time its relative value in the total above-ground part rose from 13% to 57%, due partly to the rate of uptake and partly to translocation within the plant.

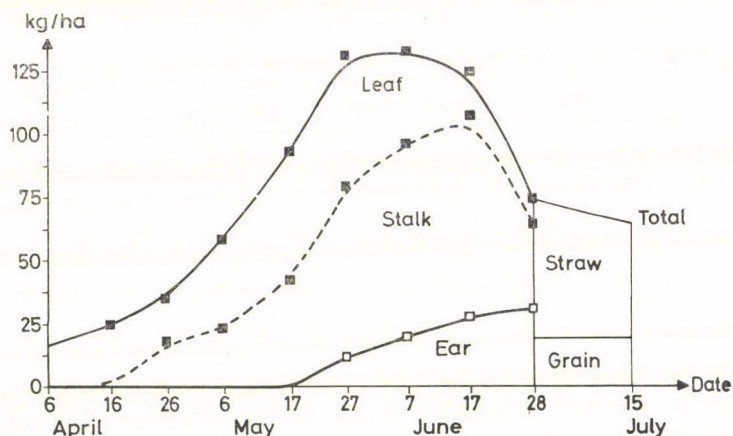


Fig. 6. Mg-uptake by winter wheat 1982

The effect of fertilization was statistically demonstrable, in the NPK treatment continuously and in the NP treatment in most cases of sampling. The PK treatment proved ineffective and so did the NK treatment on most occasions of sampling, probably on account of an antagonistic interaction between the elements.

At the time of harvesting the accumulated magnesium was stored in the grain- and straw yield at a ratio of 60—40% (Fig. 6).

### Conclusions

In a field fertilization experiment on calcareous chernozem soil, the uptake and distribution of N, P, K, Ca and Mg in the winter wheat variety "Martonvásári 8" during the vegetation period were studied. Samples were taken from 0.5 m<sup>2</sup> of each plot with 4 replications generally every 10 days from early spring until harvesting, on 10 occasions altogether. From the third occasion of sampling on the leaf and stalk, from the sixth sample taking the ear and on harvesting, the grains were separately weighed. The results of the examinations performed can be summarized as follows:

- in the leaf the uptake of N and P reached maximum on shooting, of K and Ca at the end of earing and of Mg in the phenophase of flowering. In the case of N, P and K, the rate of uptake was the lowest before harvesting, while with Mg and Ca at the beginning of shooting;
- in the stalk the uptake of N, P and Ca was the highest at the time of flowering, while of K and Mg in the phase of milky ripeness. Of N, K and Mg the rate of uptake was the lowest at the beginning of shooting, while of P and Ca later, at the end of flowering or beginning of milky ripeness;
- in the ear all the elements examined increased and reached maximum in milky ripeness;
- the proportions of the accumulated elements showed the following order of succession:

Ca, Mg, N, K, P in the leaf,  
K, N, Mg, Ca, P in the stalk,  
P, N, Mg, K, Ca in the ear,  
P, N, Mg, K, Ca in the grain,  
Ca, K, Mg, N, P in the straw,

in accordance with their physiological role;

- fertilization increased the uptake of all the elements examined compared to the control, in the total aboveground part, the stalk, the ear, and most of all in the leaf;



- the effect of fertilization was due mostly to the nitrogen-, less so to the phosphorus- and minimally to the potassium fertilizer, in the whole plant and in its parts alike.

### References

- Adorján, J. (1902): A búzaszem nitrogén felvétele (Nitrogen uptake by the wheat grain). *Kísérlet. Közlem.*, **5**, 295–304.
- Biczók Gy. — Lásztity B. (1985): Connection between the nutrient uptake by winter wheat and nutrient supply of the soil. *Zborn. Radova*, **14**, 21–28. Novi Sad.
- Biczók Gy. et al. (1985): Modelling dry matter and nutrient accumulation by winter wheat. *Agrokémia és Talajtan* **34**, Suppl. 108–119.
- Ellen, J. (1987): Effect of plant density and nitrogen fertilization in winter wheat. 1. Production pattern and grain yield. *Neth. J. Agric. Sci.* **35**, 137–153.
- Gregory, P. J. — Crawford, D. V. — Mc. Gowan, M. (1979): Nutrient relations of winter wheat. 1. Accumulation and distribution of Na, K, Ca, Mg, P, S and N. *J. Agric. Sci. Camb.* **93**, 485–495.
- Kádár, I., Csathó, P. (1985): A szuperfoszfát tartam hatásának vizsgálata őszi búza monokultúrában II. Fajlagos hatékonyság, tápelemtartalom és -felvétel, a P-előregedés vizsgálata, fenológiai megfigyelések (Long-term effect of superphosphate in winter wheat monoculture II. Specific efficiency, nutrient content and -uptake, P senescence, phenological observations). *Agrokémia és Talajtan*, **34**, 97–129.
- Kiss, E., Debreczeni, K., Pethes, J. (1985): A különböző időben adagolt nitrogén fejtárgya beépülése az őszi búza szemtermésébe. In: Búzatermesztési Kísérletek 1970–1980. (Szerk.: Bajai J. és Koltay Á.) *Incorporation of nitrogen applied by top-dressing at various times in the grain of winter wheat*. In: Wheat growing experiments 1970–1980. (Eds.: J. Bajai and Á. Koltay) 229–234. Akadémiai Kiadó, Budapest.
- Láng, I. (1960): Adatok néhány gazdasági növény ásványi táplálkozásáról réteges homokjavítás esetén (Data on the mineral nutrition of some agricultural crops in the case of stratified sand melioration). Candidate's dissertation. Budapest.
- Lásztity B. (1987): A műtrágyázás és a szervesanyag produkció dinamikája őszi búza növényben (Dynamics of nutrition and organic matter production in winter wheat). *Növénytermelés*, **36**, 105–116.
- Pethő, M. (1984): *Mezőgazdasági növények élettana* (Physiology of field crops). Mezőgazdasági Kiadó, Budapest.
- Potapov, N. G., Dézsi, L. (1954): Adatok az őszi búza ásványi táplálkozásáról szabadföldi körülmények között (Data on the mineral nutrition of winter wheat under field conditions). *Ann. Biol. Univ. Hung.*, **2**, 51–55.
- Sarić, M. (1981): Genetic specification to plant mineral nutrition. *J. of Plant Nutrition*, **3**, 743–766.
- Sarkadi J. — Krámer M. — Balla H. (1968): Die Wirkung der Stickstoffdüngung auf den NPK-Gehalt des Weizens. *Zesz. probl. Postep. Nauk Roln.* **84**, 359–364.



## THE ROOTSTOCK AS A MODIFYING FACTOR IN THE FLOWER ORGANIZATION OF THE C. 970 BESZTERCEI PLUM

D. SURÁNYI

RESEARCH AND DEVELOPMENT ENTERPRISE FOR FRUIT- AND ORNAMENTAL  
GROWING STATION Cegléd, HUNGARY

(Received 19 November 1987; accepted 19 February 1988)

Between 1977 and 1981 the author examined the flower organization properties of the C. 970 Besztercei plum clone variety on 10 myrobalan seedling stocks. The rootstock combinations greatly varied in the process of organization. The length of petiole and the pollen germination, on the other hand, showed a considerable dependence on the year. Self- and open pollination similarly fluctuated during the years of observation.

There is a relationship between the fertility (as a function) and structure of the flower. Flower with larger petals and small stigma diameters generally set better than those with the opposite characteristics. The examinations have proved that it is necessary and worth-while to place the variety research on a sounder basis, especially to concretize it by rootstock types.

The rather infrequent flower organ teratomata could be explained by the correlation of reproductive organs. Fertility can thus be influenced with the rootstock, which is of great economic importance.

**Keywords:** effect of rootstock, fertility, floral morphology, plum, sex expression

### Introduction

According to Proboeckai (1968) the rootstock is the most important environmental factor, yet, methodical investigations with plum cultivars into the effect of rootstock have rarely been carried out so far, although foreign experiences show that the rootstock induces substantial differences in cropping potential, fertility of flowers and onset of phenophases alike.

Tydemann (1957) described the most frequent and most important plum rootstocks on the basis of morphological features; according to Maurer (1939) they belong to the species *Prunus cerasifera*, *P. domestica*, *P. insititia* and *P. spinosa*. The description of their morphological characteristics is found in the book "Plum" by Tóth and Surányi (1980).

Over the last ten years, great interest has been shown in the use of rootstocks for plum cultivars. Cultivars may differ both in vegetative and reproductive characteristics depending on the rootstocks; cultivars grafted onto *Cerasus tomentosa* seedling or Pixy rootstock variety show a poor growth and this has an effect on earliness, production potential and fertility alike (cf. De Haas and Hildebrandt 1967).



"Küszvendili kék" (Joncseva 1974), "Italian Prune" (Grzyb and Jackiewicz 1978), "Pozegača" (Šoškič 1978), Stanley (Paunović 1978, Mondeska 1980), "Victoria" (Van Oosten 1977) when budded to various rootstocks, or with other varieties intergrafted, show considerable differences in growth and yield. According to Helton (1975) rootstocks may even influence infections by viruses, at least as far as the appearance of symptoms are concerned; this has been confirmed by recent observations at Cegléd. The importance of the interstock was earlier demonstrated by Tóth (1971) in nursery experiments.

Grzyb and his research team studied various aspects of the modifying role of the rootstock in many stock-scion combinations, and tried to give a possible explanation for the effect of rootstock. "Italian Prune" behaved differently on 20 rootstocks even when young (Grzyb, Jackiewicz and Czynczyk 1984, Grzyb and Zagaja 1975, Grzyb and Jackiewicz 1978). Particularly interesting is the account given by Grzyb and Zagaja (1975) of a rootstock dependent fluctuation of 11.0–31.8% in open pollination, and 22.0–44.8% with hand pollination. The self-pollination of flowers was brought into connection by Grzyb and Zagaja (1975) with the relative heights of stigma and anthers, though Tóth (1980) emphasizes in his book "Plum" too that there is no causal relation between them (cf. Surányi 1985b); however, Röder (1940), Plock (1954) and others held an opposite view.

We examined the effect of rootstock on the large-fruited and poorly self-pollinating "C. 970 Besztercei plum" cultivar from the point of view of the morphological, functional bases of fertility, using some of the methods described in our comprehensive study (Surányi 1985b).

### Materials and methods

Between 1977 and 1981, from fruit-spurs of "C. 970 Besztercei plum" trees in full blossom, we collected 100 flowers per tree. The rootstock varieties came to the Cegléd stock orchard by regional selection; data on the flower morphology of these varieties were published in two papers (Surányi 1980a, 1988).

In samples taken from trees 12–17 years old, the petiole length, pistil length, stamen number and relative stamen number were determined for each flower, the petal median (average of length and width) and stigma diameter only for every fourth flower; in 4 samples of each tree the pollen germination percentage was determined by incubation in 10% saccharose for 24 hours. The stigma diameter was measured by stage micrometer under stereomicroscope, with 10 replications (Surányi 1985b).

The rootstock effect examinations of apricot and peach (Surányi 1974), cherry (Surányi unpublished) and sour-cherry (Surányi 1985a) were regarded as empirical preliminaries. The data were statistically evaluated both for each year and on the average of the 5 years. With the observations completed (Tóth et al. 1979–1980) in the combinations chosen, self-pollination was also performed. In 1979 and 1980, the relationship between morphogenetic features and fertility was subjected to regression analysis.

From 100 flowers of each stock-scion combination, the acarpellous, apocarpous flowers and those with phylloid stamina were compared to the normal variety values. The teratomatous flowers were compared to flowers of full value, capable of functioning.

Table 1

*Fluctuation of monthly average temperature and monthly amount of precipitation between 1977 and 1981 (June–October)*

Year	June	July	August	September	October	Average
<i>Average temperature °C</i>						
1977	19.8	18.3	21.9	18.8	20.7	19.9
1978	18.3	18.8	18.5	15.5	11.6	16.5
1979	21.9	19.4	19.8	17.5	9.4	17.6
1980	18.8	19.6	19.9	15.5	11.6	17.1
1981	20.7	20.5	20.4	17.3	12.6	18.3
L. S. D. 5% = 8.29						
<i>Average precipitation, mm</i>						
1977	29.7	39.4	31.9	24.1	8.2	157.4
1978	93.9	67.0	47.2	16.1	8.2	232.4
1979	43.5	69.4	69.9	3.8	21.4	208.0
1980	101.8	44.4	35.7	22.5	61.6	266.0
1981	73.6	51.7	37.4	61.5	26.1	250.3
L S. D 5% = 85.52						

Climatic data most important from the point of view of flower bud formation are contained in Table 1, accordingly, 1977 and 1978 showed the greatest difference, as confirmed by an analysis of variance (cf. Tóth and Surányi 1980).

### Results and discussion

The major modifying effects of 10 myrobalan seedling stocks are seen in Table 2 with the lowest and highest values indicated. The rootstocks show great dissimilar differences in effect; in the case of flower organs the pistil length varied the least. In all other parameters the rootstock clones showed significant differences though not in the same measure. The fluctuation of petal median and stigma diameter is also wide; yet, the greatest variability was found in stigma diameter and pollen germination by combination, and in petiole length and pollen germination by year (Table 2).

Neither the rootstock average nor the years' average showed any considerable scatter, which is considered unusual, mainly in connection with self-pollination (Tables 1 and 3). In several cases the fertility values can be brought into close connection with morphogenetic characters, such as the petal median and the stigma diameter. The result obtained for varieties; namely, that the size of the petal and the diameter of the stigma are in close correlation seems to be true for the effect of rootstock as well (cf. Surányi 1985b), but it follows likewise from the data series of Dahl (1935) and Röder (1940). The



**Table 2**  
*Changes in the measurements of flower organs under the influence of rootstock*

Rootstock	Petiole length mm	Petal median mm	Pistil length mm	Stamen number n°	Relative stamen number n°/mm	Stigma diameter $\mu$ m	Pollen germination %
C. 168	12.4 $\pm$ 0.21	8.5 $\pm$ 0.49	15.1 $\pm$ 0.30	21.2 $\pm$ 0.07	1.41 $\pm$ 0.09	1216 $\pm$ 106	42.0 $\pm$ 3.9
C. 359	13.4 $\pm$ 0.17	9.1 $\pm$ 0.61	15.3 $\pm$ 0.28	20.5 $\pm$ 0.10	1.35 $\pm$ 0.08	1155 $\pm$ 95	46.3 $\pm$ 4.8
C. 679	13.3 $\pm$ 0.20	9.0 $\pm$ 0.37	15.1 $\pm$ 0.17	20.4 $\pm$ 0.04	1.34 $\pm$ 0.07	1188 $\pm$ 101	50.0 $\pm$ 4.4
C. 683	15.0 $\pm$ 0.24	9.4 $\pm$ 0.58	14.9 $\pm$ 0.21	19.5 $\pm$ 0.08	1.33 $\pm$ 0.09	1028 $\pm$ 79	54.3 $\pm$ 5.0
C. 722	12.3 $\pm$ 0.12	8.1 $\pm$ 0.63	14.8 $\pm$ 0.15	21.2 $\pm$ 0.12	1.42 $\pm$ 0.11	1290 $\pm$ 87	43.6 $\pm$ 3.9
C. 821	14.6 $\pm$ 0.08	9.4 $\pm$ 0.48	15.1 $\pm$ 0.33	20.3 $\pm$ 0.07	1.33 $\pm$ 0.09	1011 $\pm$ 93	50.8 $\pm$ 4.7
C. 1273	12.6 $\pm$ 0.19	8.4 $\pm$ 0.50	14.7 $\pm$ 0.25	20.3 $\pm$ 0.11	1.38 $\pm$ 0.08	1282 $\pm$ 112	48.2 $\pm$ 5.2
C. 1274	14.8 $\pm$ 0.18	9.4 $\pm$ 0.44	15.2 $\pm$ 0.30	20.5 $\pm$ 0.09	1.35 $\pm$ 0.08	1046 $\pm$ 91	55.0 $\pm$ 4.9
C. 1402	12.3 $\pm$ 0.22	8.7 $\pm$ 0.41	15.2 $\pm$ 0.19	20.6 $\pm$ 0.13	1.36 $\pm$ 0.09	1179 $\pm$ 110	46.6 $\pm$ 4.8
C. 1425	12.0 $\pm$ 0.30	8.8 $\pm$ 0.40	14.9 $\pm$ 0.26	19.7 $\pm$ 0.06	1.34 $\pm$ 0.09	1142 $\pm$ 101	57.0 $\pm$ 5.4
SD 5%	0.87	0.29	0.64	1.14	0.093	90.9	5.28
CV % for combination	5.0	5.2	1.3	0.3	4.9	8.6	9.9
for year	12.1	7.8	3.7	1.9	2.6	19.4	

Table 3

*Self-pollination and free pollination of "C. 970 Besztercei plum"  
tree in two successive years (Tóth et al., 1980)*

Rootstock	Self-pollination, %		Free pollination, %	
	1979	1980	1979	1980
C. 168	1.3	4.5	0.7	0.5
C. 359	19.7	12.9	18.6	10.8
C. 679	7.1	10.0	16.0	6.6
C. 683	21.5	17.6	33.9	17.3
C. 722	11.0	13.0	13.5	10.5
C. 821	39.2	20.2	29.0	15.0
C. 1273	14.7	15.9	7.5	23.9
C. 1274	15.7	12.9	30.5	12.6
C. 1402	18.3	14.6	18.9	7.0
C. 1425	20.5	8.7	17.5	7.0
Average	16.9	13.0	19.6	11.1

pistil length influences the fertility conditions to a lesser extent than the number of stamina and/or the amount of pollen do (Lee 1980a, 1980b), and the correlation with the relative stamen number is positively close. The importance of pollen viability is also confirmed by the positive correlation (Table 4).

Table 4

*Relationship between morphological characters and fertility  
(1979–1980)*

Independent variable	Self-pollination	Free pollination
Petal median, mm	+0.57*	+0.69**
Pistil length, mm	+0.45	+0.30
Apistilly, %	+0.19	+0.34
Polycarpy, %	+0.05	−0.15
Stigma diameter, um	−0.64**	−0.70**
Stamen number, n°	−0.70**	−0.61*
Relative stamen number, n° mm	−0.88****	−0.84****
Pollen germination, %	+0.43	+0.64**

\* p = 10%

\*\* p = 5%

\*\*\* p = 1%

\*\*\*\* p = 0.1%

In the fertility of the flower, an important role is played by the correlation of flower structure and fertility as a function, as illustrated by Fig. 1: a relatively large size of petal accompanies a lower relative stamen number,



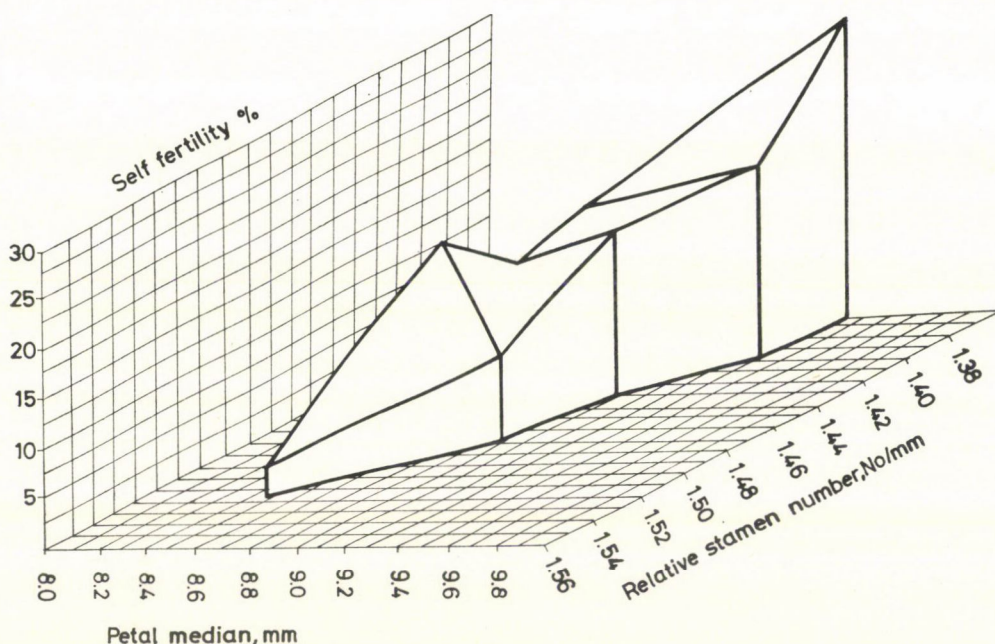


Fig. 1. Morphological components of self-fertility in "C. 970 Besztercei plum" on ten rootstocks. Self-fertility, %, Petal median, mm, Relative stamen number, no/mm

and in this case can the highest fertility be expected in the combinations of "C. 970 Besztercei plum"/myrobalan seedling.

With self-pollination managed competently, a higher fruit setting percentage is achieved than in the case of free pollination. That is, the actual problem is caused by the high degree of dichogamy and the lower intensity and shorter period of stigma secretion in plum flowers with large pistils, rather than by the reduced viability of pollen grains.

The number of abnormal flowers was negligible compared to the number of flowers examined, though sufficient to establish the correlation of reproductive organs in them (Table 5). The phylloidy of stamina and anthers characteristic of the "Besztercei plums" was not remarkable either, and could not be brought into connection with climatic factors, as was possible in the case of the varieties (cf. Surányi 1985b). We therefore only found it important to publish yearly data for the abnormal flowers.

A possible explanation for the action mechanism of rootstocks may be found in chemical factors, namely: changes in the auxin balance are favourable at some times for the gynoecium, at others for the androecium. For flowers of female character formed by the "C. 970" clone, a somewhat stronger androecium (production of a larger amount and more viable pollen) is favourable. The best hormonal conditions for forming fertile flowers are mostly given

Table 5

*Characteristics of reproductive organs in normal and teratomatous flowers  
(percentage of abnormal flowers)*

Organs	Years					Average
	1977	1978	1979	1980	1980	
<b><i>APISTILLY</i>, n =</b>	<b>0</b>	<b>16</b>	<b>10</b>	<b>0</b>	<b>0</b>	<b>16</b>
Stamen number, no						
Normal	—	19.7	20.8	—	—	20.2
Teratomatous	—	21.8	21.2	—	—	21.5
SD 5%		1.34	0.42			1.54
<b><i>POLYCARPY</i>, n =</b>	<b>3</b>	<b>2</b>	<b>2</b>	<b>2</b>	<b>1</b>	<b>10</b>
Pistil length, mm						
Normal	15.5	15.5	15.0	15.1	15.3	15.3
Teratomatous	15.0	13.5	15.3	15.0	14.9	14.7
SD 5%	0.82	0.94	1.13	0.38	0.78	0.73
Stamen number, no						
Normal	19.7	19.6	20.6	21.0	23.1	20.8
Teratomatous	19.3	19.5	20.2	20.6	24.0	20.7
SD 5%	1.15	0.94	1.02	0.67	0.48	0.78
Relative stamen number no/mm						
Normal	1.27	1.26	1.37	1.39	1.51	1.46
Teratomatous	1.28	1.44	1.32	1.38	1.61	1.41
SD 5%	0.21	0.27	0.18	0.37	0.22	0.15
<b><i>STAMINODY</i>, n =</b>	<b>26</b>	<b>20</b>	<b>50</b>	<b>51</b>	<b>15</b>	<b>162</b>
Pistil length, mm	15.2	14.9	13.5	15.1	16.1	15.0
Normal	15.3	14.8	13.6	15.1	16.9	15.1
Teratomatous	0.56	0.48	0.41	0.65	0.40	0.44
SD 5%						
Stamen number, no	19.5	20.4	19.8	20.7	21.5	20.4
Normal	18.5	18.2	19.6	20.2	19.9	19.3
Teratomatous	0.95	0.88	0.67	0.44	0.98	0.97
SD 5%						
Relative stamen number, no/mm	1.28	1.36	1.49	1.37	1.31	1.36
Normal	1.22	1.23	1.44	1.39	1.18	1.28
Teratomatous	0.32	0.19	0.20	0.18	0.31	0.63
SD 5%						



in trees on "C. 683" and "C. 821" myrobalan seedlings, as confirmed by our information on the yield per tree (cf. Surányi 1980b).

The examination of the effect of rootstock from the point of view of flower organization is very important: attention has been called to this fact mostly by Polish authors. Beside the apricot-, peach- and sour-cherry cultivars, the plum cultivars also have proved that the rootstock exercise highly diversified effects on the quantity of yield. Thus, the exploitation of this information on flower morphology may by itself have an economic importance, see also Grzyb and Zagaja, 1975.

This needs to be particularly emphasized when Hungary has arrived at a large-scale change of variety in the development of plum cultivation, and it matters a good deal whether it involves its unfavourable genetic properties or only an unfortunately chosen rootstock that produces a variety undesirable to the growers.

### References

- Dahl, C. L. (1935): Morphological studies of plum flowers. *Meded. Fruchtdl. Försök*, **38**, 1-93.
- Grzyb, Z. S., A. Jackiewicz, (1978): The influence of sinterstocks on the growth and yield of Italian Prune cultivar. *Acta Hort. Hague* **74**, 185-189.
- Grzyb, Z. S., A. Jackiewicz, A. Czynczyk, (1984): Results of the 18-years evaluation of rootstocks for Italian Prune cultivar. *Fruit Sci. Rep.* **11**, 99-104.
- Grzyb, Z. S., S. W. Zagaja, (1975): The influence of various rootstocks on the morphological changes in Italian Prune flowers. *Acta Hort. Hague* **48**, 7-11.
- Haas, P. G., De W. Hildebrandt, (1967): *Die Unterlagen und Baumformen des Kern- und Steinobstes*. Ulmer, Stuttgart.
- Helton, A. W. (1975): Effect of cultivar and rootstock on incidence of viral and nonviral symptoms in PRSV-infected *Prunus domestica* L. *Phytopath.* **65**, 539-541.
- Joncseva, M. (1974): Studies on semi seed-propagated rootstocks for plum cultivar Kűszten-dilcszka szinja. *Grad. Loz. Nauka* **11**, 15-30.
- Lee, C. L. (1980a): Pollenkeimung, Pollenschlauchwachstum und Befruchtungsverhältnisse bei *Prunus domestica*. I. Pollenkeimung *in vitro* und *in vivo*. *Gartenbauwiss.* **45**, 228-235.
- Lee, C. L. (1980b): Pollenkeimung, Pollenschlauchwachstum und Befruchtungsverhältnisse bei *Prunus domestica*. II. Pollenschlauchwachstum im Griffel. *Gartenbauwiss.* **45**, 241-248.
- Maurer, E. (1939): *Die Unterlagen der Obstgehölze*. Parey, Berlin.
- Mondeszka, P. (1981): Izzledvanija vörhu korevonata sziszte szlivovü dörvata, priszadeni na razlicsni podlozski. *Grad. Loz. Nauka* **18**, 28-38.
- Oosten, H. J. Van (1977): Rootstocks and interstocks of pit and stone fruits. *Ann. Rept.* **1976**. Res. Sta. Fruit Growing Wilhelminadorp, 22-23.
- Paunovic, S. A. (1978): The effect of sootstocks on yield and quality of prunes cvs Požegača and Stanley. *Acta Hort. Hague* **74**, 175-183.
- Plock, H. (1954): Die Befruchtung bei unseren Obstarten. *Obst- und Gartenbau* **3**, 68-69.
- Probocskai E. (1968): Faiskola (Nursery). Mezőgazdasági, Budapest.
- Röder, K. (1940): Sortenkundliche Untersuchungen an *Prunus domestica* L. *Kühn-Archiv* **54**, 1-133.
- Soskič, A. (1978): Uticaj podloge na visinu prinosa i kvalitet plodova sljive "Požegače". *Savr. Poljopr.* **26**, 87-94.
- Surányi D. (1974): Influence of the flower-organization of *Prunus* species by rootstocks. *Bot. Köz.* **61**, 117-120.
- Surányi D. (1980a): Data to flower morphology of cherry plums. *Bot. Köz.* **67**, 301-306.

- Surányi D. (1980b): *The balance of male and female sex in the Prunoidea flowers*. In the book "Gyümölcsfajták virágzásbiológiája és termékenyülése", Mezőgazdasági, Budapest.
- Surányi D. (1985a): Change of sex expression of sour cherry varieties by rootstocks. *Acta Agron. Hung.* **34**, 233-242.
- Surányi D. (1985b): *Flower structure of historical and cultivated plums, connection of morphological remarks to self-fertility*. Dissertation of Hungarian Scientific Academy, Budapest.
- Surányi D. (1988): Floral morphological investigations on selected local plums. *Acta Hort. Hague* **224**, 461-468.
- Tóth E. (1971): Result of interstocks for plum in nursery. *Szőlő- és Gyüm. term.* **6**, 165-174.
- Tóth E., D. Surányi (1980): *Plum*. Mezőgazdasági, Budapest.
- Tydemann, H. M. (1957) A description and classification of certain plum rootstocks. *Ann. Rept. London* 1956, 75-80.





## RESPONSE OF WHEAT (*TRITICUM AESTIVUM* L). TREATED WITH CYCOCEL UNDER WATER STRESS CONDITIONS

M. YASIN ASHRAF,<sup>1</sup> N. A. BAIG<sup>2</sup> and F. BAIG<sup>2</sup>

<sup>1</sup> ATOMIC ENERGY AGRICULTURAL RESEARCH CENTRE, TANDOJA, PAKISTAN

<sup>2</sup> DEPT. OF BOTANY, UNIVERSITY OF AGRICULTURE FAISALABAD, PAKISTAN

(Received 20 July 1987; accepted 3, September 1987)

The response of wheat cultivar treated with cycocel (CCC) under water stress conditions was studied at the Agriculture University Farm Faisalabad. The seeds were soaked in 0, 600, 1000, 1400 ppm CCC, after which they were sown in field under three levels of water i.e. (1) drought, (2) two irrigation, (3) four irrigation. The CCC treatments significantly decreased plant height, and number of sterile spikelets at all levels of water treatments. On the other hand the number of tillers, number of ears per plant, ear length, number of grains per ear, thousand grain weight and yield per plot were increased. Whereas under different water levels there was no effect on the number of sterile spikelets per ear, the effect on the above-mentioned characters were identical to that of CCC.

**Keywords:** cycocel, water stress, yield components

### Introduction

Water available for irrigation is usually limited, but its efficient use is important. Irrigating crops to avoid yield reduction by water stress may not be the most rational use of water in the economic sense. An alternative strategy is to use less water per unit area and irrigate more land to compensate for lower yield and give greater returns (Constable and Hearn 1980).

The use of cycocel (2-chloroethyl trimethyl ammonium chloride) has better results under limited water supply and drought conditions (Baig, 1970). According to Giri and Singh (1984) cycocel enables plants to withstand the drought condition, and to consume water more economically. It also induces xeromorphic characters, reduces plant height and the adverse effects of salinity (Kazim and Mohsin, 1980). Cycocel treatment enlarges the root system of plants and allows more tillers to survive when the soil is dry (Adler, 1966).

The effects of water deficits and cycocel on growth, yield, yield components and water use efficiency of wheat have not been studied extensively under the stressful conditions in the semiarid area.

We have attempted to study the response of wheat cultivar L. U. 26S (*Triticum aestivum* L.) under water stress conditions.



## Materials and methods

The experiment consisted of three irrigation treatments and four cycocel treatments in a split plot design with three replications. Irrigation treatment occupied main plots ( $12 \times 64$  sq feet) and cycocel treatment occupied sub plots ( $12 \times 16$ ). Sub plots contained ten rows of wheat at a distance of one foot apart.

### *Cycocel treatment*

The seeds of wheat cv. L. U. 26 S were soaked in 0, 600, 1000 and 1400 ppm cycocel separately. After 24 hours seeds were sown in their respective sub plots which were selected randomly in each replication.

### *Irrigation treatment*

This experiment consisted of three irrigation treatments. The main plots selected for irrigation treatment first (D) were not irrigated during the wheat growing season (drought or water deficit treatment). Similarly the plot selected for irrigation treatment second (L) and third (N) were irrigated two and four times respectively, in the whole season. At the end of the experiment, the plants were harvested and plant height, number of tillers, ear heads per plant, ear length, number of grains per ear, 1000 seed weight, yield per plot, number of sterile spikelets per ear were recorded, and the results were analysed statistically in which Duncan's multiple range test was conducted (Steel and Torrie, 1960).

### *Irrigation schedule*

Irrigation level I (D)	The plots of this treatment were not irrigated during the wheat growing season.
Irrigation level II (L)	The plots of this treatment were given first irrigation at vegetative stage and second at flowering stage.
Irrigation level III (N)	The plots of this treatment were given first irrigation at early vegetative stage, third at preflowering, and fourth at earing.

## Results and discussion

Cycocel as well as water stress reduced the plant height of wheat (Table 1). The effect of cycocel was much more pronounced at the first irrigation level (D) and 1400 ppm cycocel than at second (L) and third (N) irrigation levels. At the first irrigation level, reduction in plant height by cycocel amounted to 7.02% while at irrigation level second (L) and third (N) reduction was 6.80% and 6.95% respectively. Reduction of water supply from four irrigation to no irrigation shortened the plant by 1.35%. Similar results were reported by Bechyne (1982) Braun and Wild (1985), Wooley (1982) and Herbert (1982). Who found that the inhibitory effect of cycocel can be interpreted as an inhibition of gibberellin biosynthesis.

The first irrigation level (D) led to the production of tillers at a late stage of growth. Their height was reduced in the case of the treatments with cycocel but the number of tillers per plant was more in those treated with cycocel at all irrigation levels (D). The plants treated with cycocel did not only shows a tendency of an increase in the number of ear heads per plant but also a decrease in number of sterile spikelets per ear. The results were

Table 1

*Effect of water stress and cycocel on growth, yield and yield components of wheat*

Irrigation Treatment	Control	600 ppm CCC	1000 ppm CCC	1400 ppm CCC	Mean
<i>Plant height in cm</i>					
D(drought)	57.67	55.37	54.50	53.62	55.29b
L(two irrigation)	57.92	55.60	54.80	53.98	55.57b
N(four irrigation)	58.64	56.13	54.85	54.56	56.04a
Mean	58.08a	55.7b	54.72c	54.05d	
<i>No. of tiller per plant</i>					
D	8.12	9.39	9.97	9.82	9.33 b
L	8.23	9.41	10.17	9.93	9.44 b
N	8.61	10.14	10.74	10.41	9.98 a
Mean	8.32 d	9.65 c	10.31 a	10.05 b	
<i>No. of ear per plant</i>					
D	8.10	9.34	9.90	9.80	9.29 b
L	8.20	9.40	10.02	9.87	9.37 b
N	8.50	10.05	10.53	10.07	9.79 a
Mean	8.27 c	9.60 b	10.15 a	9.91 a	
<i>Length of ear head cm</i>					
D	12.67	13.15	13.56	13.50	13.22 a
L	12.67	13.23	13.67	13.50	13.28 a
N	12.68	13.25	13.70	13.53	13.29 a
Mean	12.67 b	13.21 d	13.64 a	13.51 a	
<i>No. of grain per ear</i>					
D	57.20	63.93	64.17	63.86	62.29 b
L	58.23	64.00	64.18	64.07	62.62 a
N	58.77	64.07	64.23	64.23	62.83 a
Mean	58.07 b	64.00 a	64.19 a	64.05 a	
<i>No. of sterile spikelets per ear</i>					
D	0.69	0.33	0.16	0.33	0.38 a
L	0.67	0.30	0.10	0.31	0.35 a
N	0.68	0.31	0.12	0.28	0.35 a
Mean	0.68 a	0.31 b	0.13 c	0.31 b	
<i>1000 grain weight (gm)</i>					
D	46.90	50.97	52.43	51.46	50.44 b
L	46.95	50.99	52.92	52.00	50.72 ba
N	48.86	51.08	52.96	52.06	51.24 a
Mean	47.57 c	51.01 b	52.77 a	51.84 a	
<i>Yield per plant (gm)</i>					
D	1233.30	1606.70	1700.00	1559.3	1524.83 b
L	1302.00	1713.30	1790.00	1700.3	1626.40 a
N	1423.3	1719.60	1803.3	1708.4	1663.65 a
Mean	1319.53 c	1679.86 ba	1764.43a	1656.0 b	

Any two means sharing same letters are non-significant at 5% level.



favourably influenced by cycocel especially, under high water stress (no irrigation) (Fraggatt et al. 1982). Nay and Tabi (1982) and Pikush and Sakhrov (1982), reported that the application of cycocel inhibited auxin effect in the apical part of the plant, thus decreasing apical dominance and stimulating the release of nodes and lateral branches. Due to this the number of tiller and number of ear heads per plant increased.

Ear length was not affected appreciably by cycocel (Table 1). Also there was not much influence on the number of grains per ear except at irrigation level (D). The ear length and number of grains per ear was highly increased by cycocel, especially at 1000 ppm cycocel (10% in number of grain per ear and 7.65% in ear length). In first irrigation level (D) the ear length and number of grains per ear were 7.02% and 12.18% higher than in untreated plants. This indicates that, in this case of water stress, the plant treated with cycocel showed better conditions for fructification or cell division after fructification. Similar reports were given by Kan and Wasti (1980). Giri and Singh (1984, 1982) working on wheat have also found a tendency for an increase in ear length and number of grains per ear in consequence of cycocel and water stress treatments.

Plants treated with cycocel showed a tendency toward an increase in the 1000 grain weight and total yield per plot. In irrigation level "D" and "L", this increase was significant 11.79% and 12.69% in 1000 grain weight than in irrigation (N) (8.0%). Similarly, the yield per plot was 37.84% more in plants treated with cycocel than untreated at irrigation level "D" and 37.48% at irrigation level "L" while it increased 26.70% at irrigation level "N" irrigation which clearly showed that cycocel exerted a favourable influence on 1000 grain weight and total yield per plot, when plants were under high water stress. The best results were obtained by 1000 ppm cycocel at all irrigation levels. This agrees with the observation of Giri and Singh (1984, 1982). Who found that in seeds of winter wheat treated with cycocel and water stress conditions, the 1000 grain weight and yield per plot were increased.

### References

- Adler, H. (1966): Investigations into yield structure of winter wheats treated with CCC and with split applications of N. *Bodenkulture*, **17**, 165-73.
- Baig, F. (1970): The effect of (2-chloroethyl) trimethyl ammonium chloride (CCC) and gibberellic acid on the growth of *Helianthus annuus* grown at different moisture regims. *Pak J. Agric. Sci.* **7** (2), 42-53.
- Bechyne, M. (1982): The response of spring oilseed crops to CCC treatment. *Sbornik Vysoke Skoly Zemedelske V Proze, Fakulta Agronomicka A.* **36**, 179-195.
- Braun, P., Wild, A. (1985): The influence of brassionsteroid on growth and parameters of photosynthesis of wheat and mustard, *Z. Pflanzen Physiol.* (In press.)
- Constable, G. a., Hear, A. B. (1980): Irrigation for crops in sub humid environment 1. Effect or irrigation on the growth and yield of soybeans. *Irrigation Sci.* **2**, 1-12.
- De, R., Giri, G., Saran, G., Singh, R. K. (1982): Chaturved. Modification water balance of dryland wheat through the use of chlormequat chloride. *J. Agric. Sci. Comb.* **98**, 593-597.

- Fraggatt, P. J., Tomas, W. D., Batch, J. J. (1982): *The value of lodging control in winter wheat as exemplified by the growth regulators*. Monograph. British Plant Growth Regulator Group 7, 71-81.
- Giri, G., Singh, R. R. (1984): Water consumption and economics of wheat production as influenced by mulch and transpiration suppressants under drylands. *India J. Agro.* **29** (2), 173-178.
- Herbert, C. D. (1982): *Growth regulation in cereals chance or design*. In chemical manipulation of crop growth and development. London U. K. Butterworths London, U. K. pp. 315-327.
- Kazim, A. A., Mohsin, H. (1980): Effect of application of CCC and saline water on growth and yield characteristics of tomato plants. *Mesopotamia Journal of Agriculture* **15** (2), 157-167.
- Khan, K. A., Wasti, A. K. (1980): Influence of CCC on the growth and development of wheat. *Pak. J. Scient. Indust. Res.* **26** (2), 239-242.
- Nagy, M., Tabi, Z. (1982): Effect of chloro choline chloride on the amount of diffusible gibberellins in bean plants. *Biochem. Physiol. Pflanzen.* **177**, 725-728.
- Pukish, G. R., Sakharov, V. D. (1982): Application of Tur (CCC) for increasing yield of irrigated winter wheat. *Khimiya V Sels' Kom Kohzyaistue* **20** (J), 42-45.
- Schneider, A. D., Musick, J. T., Dusek, D. A. (1969): Efficient wheat irrigation with limited water. *Trans ASAE* **12**, 23-26.
- Steel, R. G. D., Torrie, J. H. (1960): *Principle and procedures of statistics*. McGraw Hill company Inc. New York, **183**, 28-286.
- Wooley, E. F. (1982): *Performance of current growth regulators in cereals*. Monograph, British Plant Growth Regulator Group, 7, 44-50.
- Zaher, A., Foad, M. K. Shaarawi, A. El., El-Fouly, M. M. (1973): Morphological and anatomical modifications in wheat after treatment with chlormequat chloride (CCC) *Egyptian. J. Bot.* **26** (1-3), 125-136.





## INTERACTIVE EFFECTS OF SALINITY AND PHYTOHORMONES ON GROWTH AND PLANT WATER RELATIONSHIP PARAMETERS IN MAIZE AND SAFFLOWER PLANTS

A. F. RADI,\* M. M. HEIKAL,\* A. M. ABDEL-RAHMAN\*\* and  
B. A. A. EL-DEEP\*\*

\*DEPARTMENT OF BOTANY, FACULTY OF SCIENCE, ASSIUT UNIVERSITY, EGYPT

\*\*DEPARTMENT OF BOTANY, FACULTY OF SCIENCE, ASSIUT UNIVERSITY SOHAG, EGYPT

(Received 10 July 1987; accepted 23 November 1987)

The effect of various levels of NaCl salinization on seed germination, growth and plant-water relationships of maize and safflower plants was investigated. The role of  $GA_3$  and IAA in modifying the salt-stress induced changes was also studied.

The percentage germination and water content of stressed seeds were significantly reduced, but this inhibitory effect was eliminated by seed presoaking with  $GA_3$  or IAA.

Salinity induced a considerable reduction in transpiration rate and stomatal frequency, but seed presoaking with  $GA_3$  or IAA induced a significant increase in these plant-water relationships.

The dry matter yields of salt affected plants were significantly reduced. Such inhibitory action was decreased by treating the seeds with  $GA_3$  or IAA particularly in the case of maize plants.

**Keywords:** *Carthamus tinctorius* L., safflower, *Zea mays* L., maize, salinity, water relations

### Introduction

A knowledge of the physiological effects of soil salinity and exogenously applied growth hormones on seed germination, plant growth and the relevant vital activities, provides a fundamental basis for the intelligent management of plant life for the good of mankind. In the majority of plants, germination of seeds is greatly retarded and seedling survival is difficult under saline conditions (Maftoun and Sepaskhah, 1978; Stout et al., 1980 and Khan and Naqvi, 1984). In addition, the response of plant growth to salinization treatment has been investigated. In this respect, some investigators reported a general reduction in plant growth (Stewart et al., 1976; Verasan and Phillips, 1978; Ahmed et al., 1980a, Ralph et al., 1984; Nerson and Paris, 1984 and Patil et al., 1984) while others recorded a promotion rather than inhibition in the growth of some salinized plants (Bernstein, 1975; Ahmed et al., 1980b and Heikal et al., 1980 and 1981b). According to Kessler (1961), Shah and Loomis (1965), the major effect of salinity in the root environment was attributed to a reduced hormone delivery from root to leaves which could induce an inhibition of crop growth. Attempts have been made to overcome the



growth suppression resulting from salinity and other factors such as genetic deficiencies or restrictive light and temperature conditions (Nieman and Bernstein, 1959). In this respect, presoaking seed with optimal concentration of certain phytohormones has been shown to be beneficial to growth and yield of some plants grown under saline conditions (Kaufmann and Ross, 1970; Gary and Srivastava, 1970; Darra et al., 1973, Chhipa and Lal, 1978, and Shaddad and Heikal, 1982).

During the vegetative growth phase, salinity has been recognized as one of the factors which induce significant changes in plant-water relationships. In this respect, many investigators recorded a reduced transpiration rate with the rise of salinity level (Tal and Gavish, 1973; Bozcuk, 1975; Stewart et al., 1976; Ahmed et al., 1980a, Eshel, 1985; and Melzack et al., 1985). This reduction was found to be associated with reduced leaf surface area. Also, the stomatal number and movement were found to play an additional role (Bozcuk, 1975 and Ahmed et al., 1979a). In accordance with this, Waisel (1972) and Shaddad and Heikal (1981) reported that the number, size and movement of stomata were considerably affected with the rise of NaCl concentration in the nutritive medium.

The changes in transpiration rate due to salinization treatments lead mostly to concomitant changes in plant water content (Wong and Jager, 1978; Adams et al., 1978; and Zidan, 1979). Poljakoff-Mayber and Gale (1975) attributed the increase in water content to an increase in abscisic acid concentration, which in turn induces stomatal closure, leading to an increase in leaf-thickness, or succulence.

The present work was undertaken to study the effect of various levels of salinity on seed germination, growth and plant-water relationships of maize (*Zea mays* L.) and safflower (*Carthamus tinctorius* L.) plants. Because of the various biological activities of GA<sub>3</sub> and IAA, their role in modifying the salt-stress induced changes was also investigated.

### Materials and methods

Preliminary tests were conducted to determine the optimum hormone concentration and optimum period of presoaking and drying which are effective in counteracting the effect of salt-stress on seed germination. The optimum conditions reached from these tests are shown in the following scheme:

Test plant	The optimum conditions					
	GA <sub>3</sub>			IAA		
	Conc, ppm	Period of presoaking	Period of drying	Conc, ppm	Period of presoaking	Period of drying
Maize	100	3 hours	one day	50	3 hours	3 days
Safflower	100	5 hours	7 days	100	3 hours	3 days

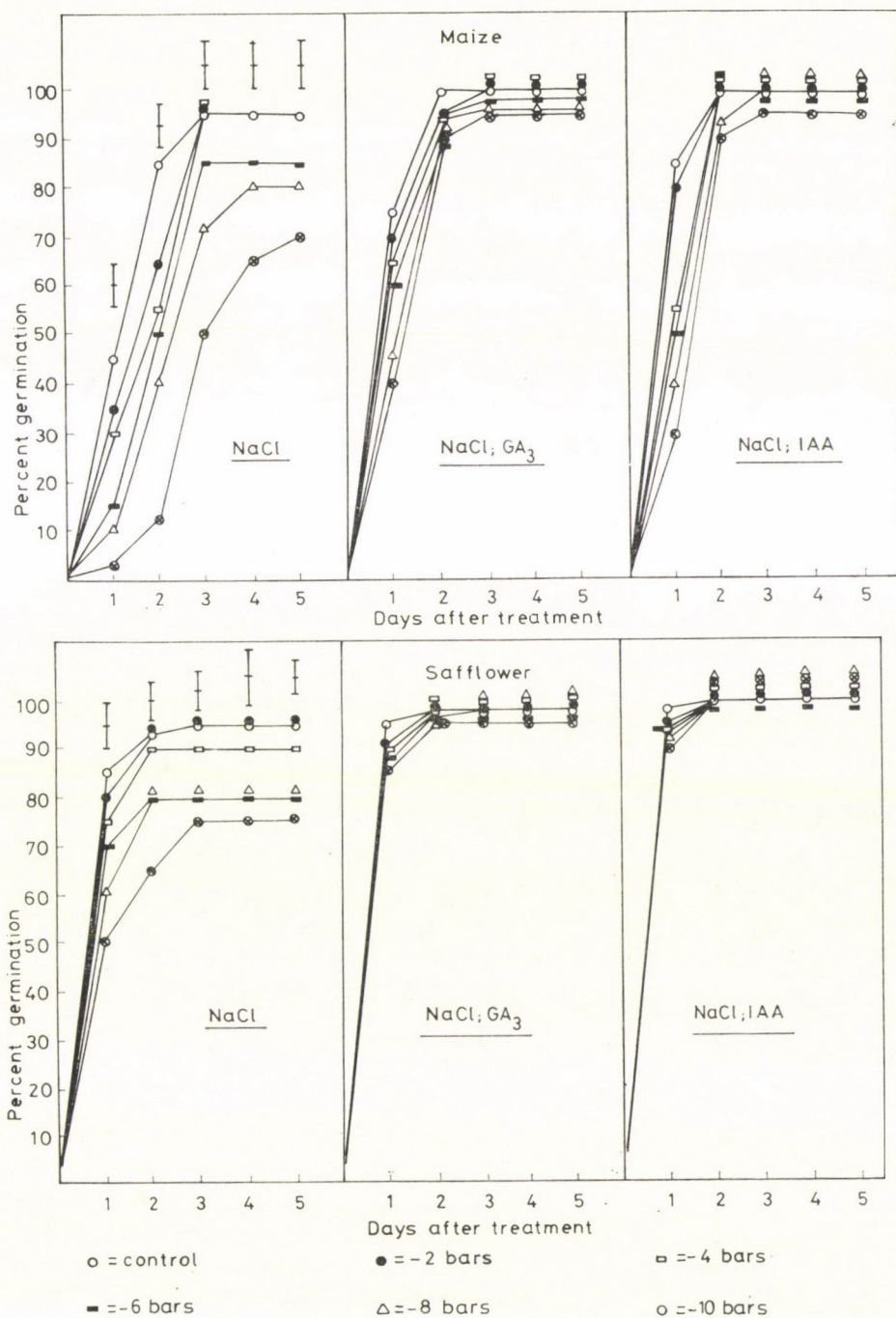


Fig. 1. Effect of seed presoaking with GA<sub>3</sub> and IAA on percent germination of maize and safflower seeds stressed with different levels of sodium chloride. ○ = control, ● = −2 bars, □ = −4 bars, ■ = −6 bars, △ = −8 bars, ▲ = −10 bars



Using NaCl the osmotic potential levels were chosen at 0 (control), -2, -4, -6, -8 and -10 bar. Twenty-five seeds were placed on observant pads in Petri dishes to which 25 ml of the experimental saline solution was added. The Petri dishes were wrapped in two layers of aluminium foil and incubated at 25 °C. Seeds were considered to be germinated after the radicle emerged from the testa. To evaluate the interactive effect of salt-stress and GA<sub>3</sub> or IAA on seed germination, the seeds of the test plants were treated with the experimental phytohormones, air dried and germinated under the effect of various levels of salinity. The percentage germination was followed daily for a period up to 5 days.

The water content, soluble carbohydrate and soluble nitrogen of the germinated seeds were determined. The anthrone sulphuric acid method (Fales, 1951; Schlegel, 1956 and Badour, 1959) was used for determining the carbohydrate content. For nitrogen determination, the micro-Kjeldahl method was employed (Paech and Tracey, 1956).

To follow up the interactive effect of salinity and phytohormones on seedling growth, the phytohormone treated seeds were placed between folded paper towels in a beaker containing 80 ml of the salinized solution and incubated at 25 °C for 10 days in darkness. The length of shoots and primary roots in the case of maize, and length of hypocotyl and roots in that of safflower, were recorded.

For determining plant growth and plant-water relationship parameters, seeds of the test plants, after being presoaked and air dried were sown in plastic pots containing 2 kg air dried soil (sand/clay 1 : 1 v/v). Stress levels were at -2, -4, -6, -8 and -10 bar in the case of maize and at -3, -6, -9 and -12 bar in that of safflower. Plants were irrigated every other day with a salinized nutrient, Pfeffer's solution, for two weeks. Thereafter, the test plants were irrigated every other day with N/10 non-salinized Pfeffer's solution. The soil moisture content was kept near the field capacity.

Table 1

*Changes in water content (g/g dry weight), soluble carbohydrate and soluble nitrogen (mg/g dry weight) of maize seeds treated with GA<sub>3</sub>, IAA and stressed with different levels of sodium chloride for three days*

Treatment	Water content	Soluble carbohydrates	Soluble nitrogen
Control	0.83	42.10	6.85
-2 bar	0.89*	44.55*	10.69**
-4 bar	0.63**	31.59**	8.77**
-6 bar	0.55**	25.11**	7.02
-8 bar	0.49**	17.01**	6.56
-10 bar	0.47**	14.58**	6.12*
Control + GA <sub>3</sub>	1.03**	48.60**	12.50**
-2 bar + GA <sub>3</sub>	0.89	47.36**	11.05
-4 bar + GA <sub>3</sub>	0.81**	48.69**	10.08**
-6 bar + GA <sub>3</sub>	0.72**	46.70**	9.17**
-8 bar + GA <sub>3</sub>	0.68**	46.56**	7.96**
-10 bar + GA <sub>3</sub>	0.51	43.59**	8.77**
Control + IAA	0.95**	65.12**	7.26
-2 bar + IAA	0.96**	50.62**	7.46**
-4 bar + IAA	0.71**	42.77**	7.86**
-6 bar + IAA	0.65**	35.17**	5.44**
-8 bar + IAA	0.58**	35.96**	5.44**
-10 bar + IAA	0.55**	23.33**	6.75*
L.S.D. at 5%	0.05	2.35	0.60
L.S.D. at 1%	0.07	3.15	8.81

\* Significant differences.

\*\* Highly significant differences as compared with the control.

Table 2

*Changes in water content (g/g dry weight), soluble carbohydrates and soluble nitrogen (mg/g dry weight) of safflower seeds treated with GA<sub>3</sub>, IAA and stressed with different levels of sodium chloride for three days*

Treatment	Water content	Soluble carbohydrates	Soluble nitrogen
Control	3.35	67.23	7.25
-2 bar	2.63**	59.94**	7.26
-4 bar	1.82**	57.51**	6.71
-6 bar	1.40**	29.16**	6.15**
-8 bar	1.32**	21.06**	6.15**
-10 bar	1.38**	21.06**	6.47*
Control+GA <sub>3</sub>	3.84*	78.84**	9.03**
-2 bar+GA <sub>3</sub>	3.30**	81.61**	11.99**
-4 bar+GA <sub>3</sub>	2.41**	61.56**	10.60**
-6 bar+GA <sub>3</sub>	1.85*	39.69**	8.77**
-8 bar+GA <sub>3</sub>	1.90**	37.26**	7.66**
-10 bar+GA <sub>3</sub>	1.55	33.21**	6.75
Control+IAA	1.20**	26.63**	6.75
-2 bar+IAA	1.15**	28.54**	9.68**
-4 bar+IAA	0.84**	39.69**	9.96**
-6 bar+IAA	0.94*	45.36**	8.77**
-8 bar+IAA	0.83*	43.74**	7.66**
-10 bar+IAA	0.79**	41.31**	6.75
L.S.D. at 5%	0.42	2.84	0.69
L.S.D. at 1%	0.57	3.80	0.92

\* Significant differences.

\*\* Highly significant differences as compared with the control.

Transpiration rate was measured as described by Bozcuk (1975). Young seedlings were transferred to aerated 1/5 strength Pfeffer's nutrient solution for 5-9 days until several leaves were fully expanded. Then they were transferred to a 250 ml conical flask covered with aluminium foil containing 200 ml of the experimental salinized nutrient solutions (0 to -10 bar for maize and 0 to -12 bar for safflower). In order to prevent evaporation, the flasks were sealed with paraffin wax. The transpired water was then determined by weighing the plants together with the flask and experimental salinized nutrient solution at daily intervals for 15 days. After each weighing the level of the culture solution was brought to the initial level by an addition of the nutrient solution. The transpiration rate was calculated as g per unit leaf area per day (g/dm<sup>2</sup>/day) after subtraction of the amount of water lost from a control flask kept under the same conditions, except that no plants were cultivated in it.

For stomatal frequency determination, direct microscopic measurements were carried out on strips taken from the leaves under investigation and immediately immersed in absolute alcohol for fixation and preservation. The number of stomata per mm<sup>2</sup> (stomatal frequency) on upper and on lower epidermis was determined using the square ocular micrometer.

At the end of the experimental period (45 days for maize and 60 days for safflower) the leaf area (dm<sup>2</sup>/plant) and dry matter yield were determined.

## Results and discussion

The seed germination of the test plants was generally reduced by salinization and the reduction was more pronounced with the rise of salinizing agent (Fig. 1).



It seems that this reduction in seed germination is a general response to salinization treatments as realized by Parmer and Moore (1968); Ghorashy and Kheradnam (1972); Sionit et al. (1973); Maftoun and Sepaskhah (1978); Stout et al. (1980); Kan and Naqvi (1984) and Jeffrey and Critchley (1985). The promotive effect of  $GA_3$  and IAA on germination of salt-stressed seeds of the test plants agrees with the results obtained by other authors (Darra et al., 1973; Boucaud and Unger, 1976; Stout et al., 1980; Heikal et al., 1982a; and Lin, 1985). This promotive effect was associated with an increase in water uptake (Tables 1 and 2), which again confirms the suggestion that the beneficial effects of hormones is ascribed to their role in increasing water absorption (Darra et al., 1973).

There is also a considerable reason to believe that the alteration in the production of soluble carbohydrates and soluble nitrogen in the germinated seeds may be one aspect of the role of salinity on the enzymatic activity in the overall phenomenon of seed germination. The obvious increase in the

Table 3

*Effect of seed presoaking with  $GA_3$  and IAA on the growth of seedlings of maize and safflower stressed for 10 days with different levels of sodium chloride*

Treatment	Maize		Safflower	
	Shoot length (cm)	Root length (cm)	Hypocotyl length (cm)	Root length (cm)
Control	9.61	15.73	9.34	14.40
— 2 bar	13.89**	20.70**	10.67**	18.67**
— 4 bar	12.43**	19.96**	8.22**	14.29
— 6 bar	10.06	14.23*	6.36**	9.68**
— 8 bar	7.35**	10.95**	5.46**	9.55**
— 10 bar	5.32**	6.75**	3.69**	3.19**
Control+ $GA_3$	14.74**	19.85**	10.56**	14.30
— 2 bar+ $GA_3$	19.94**	22.44*	11.83**	19.25
— 4 bar+ $GA_3$	13.69*	21.45*	9.66**	16.83**
— 6 bar+ $GA_3$	10.40	18.87**	8.18**	10.93*
— 8 bar+ $GA_3$	9.91**	15.37**	6.44**	10.92**
— 10 bar+ $GA_3$	6.83**	8.42*	4.69**	3.37
Control+IAA	12.18**	15.51	8.22**	6.64**
— 2 bar+IAA	14.89*	20.60	7.02**	6.67**
— 4 bar+IAA	12.19	14.53**	5.85**	5.32**
— 6 bar+IAA	11.13*	11.58**	4.17**	1.87**
— 8 bar+IAA	8.53*	8.41**	2.52**	1.04**
— 10 bar+IAA	6.56*	6.55	1.47**	0.80**
L.S.D. at 5%	0.99	1.46	0.73	0.95
L.S.D. at 1%	1.33	1.95	0.98	1.27

\* Significant differences.

\*\* Highly significant differences as compared with the control.

Table 4

*Effect of seed presoaking with GA<sub>3</sub> and IAA on leaf area (dm<sup>2</sup>/plant), transpiration rate (g/dm<sup>2</sup>/day) and stomatal frequency (number of stomata/mm<sup>2</sup>) of maize plants stressed for 15 days with different levels of sodium chloride*

Treatment	Leaf area	Transpiration rate	Stomatal frequency	
			Upper epidermis	Lower epidermis
Control	1.85	4.39	162	266
— 2 bar	1.64*	3.45**	140**	244**
— 4 bar	1.49**	2.26**	103**	207**
— bar	1.24	3.17**	88**	177**
— bar	0.95**	2.66**	66**	140**
— 10 bar	0.57**	1.98**	44**	10**
Control+GA <sub>3</sub>	1.89	4.69*	199**	295**
— 2 bar+GA <sub>3</sub>	1.82*	3.92**	169**	244
— 4 bar+GA <sub>3</sub>	1.77**	3.84**	147**	207
— 6 bar+GA <sub>3</sub>	1.29	3.72**	140**	199**
— 8 bar+GA <sub>3</sub>	0.97	2.74	103**	147
— 10 bar+GA <sub>3</sub>	0.58	2.06	51	111
Control+IAA	2.26**	5.07**	199**	280
— 2 bar+IAA	1.82*	4.72**	169**	258
— 4 bar+IAA	1.59	4.36**	147**	222
— 6 bar+IAA	1.32	4.24**	133**	199**
— 8 bar+IAA	0.95	3.43**	125**	177**
— 10 bar+IAA	0.58	2.87**	80**	133**
L. S. D. at 5%	0.18	0.28	13.92	15.22
L. S. D. at 1%	0.24	0.38	18.65	19.58

\* Significant differences.

\*\* Highly significant differences as compared with the control.

concentration of soluble carbohydrates in the stressed seeds as a result of presoaking with GA<sub>3</sub> (Tables 1 and 2) supports the opinion of Ikuma and Thimann (1963), who were led to conclude that GA<sub>3</sub> promotes amylase activity which supplies the monosaccharides for embryo respiration. Also, seed presoaking with GA<sub>3</sub> was accompanied by an increase in the concentration of the soluble nitrogen fractions in the test seeds (Tables 1 and 2). From this, it can be suggested that GA<sub>3</sub> not only promotes amylase activity but also acts as an activator of some hydrolases such as proteolytic enzymes. On the other hand, IAA induced a significant reduction in the contents of soluble nitrogen of saltstressed seeds (Tables 1 and 2).

The lengths of the root and shoot of maize seedlings were significantly increased at the lower levels of salinization (—2 and —4 bar). Thereafter, the values of these growth parameters decreased as the level of salinity increased (Table 3). In case of safflower the lengths of the hypocotyl and root were significantly decreased with the rise of salinization levels, except at the lowest level (—3 bar) where these growth parameters were significantly



increased (Table 3). Presoaking with  $GA_3$  induced a significant increase in seedling growth parameters of the two test plants, at all chosen stress levels (Table 3). Presoaking with IAA generally retarded the root growth of maize seedlings and hypocotyl and root growth of safflower seedlings at various levels of salinization. On the other hand, the length of shoots in salinized maize seedlings was generally increased under the effect of IAA.

The reduction in stomatal frequency and rate of transpiration of the test plants at certain concentration of NaCl (Tables 4 and 5) accords with the results obtained by some other authors (Meiri and Poljakoff-Mayber, 1970; Tal and Gavish, 1973; Jensen, 1975 and Ahmed et al., 1980a). Gale et al. (1967) and Bozcuk (1975) attributed such inhibited transpiration to the partial closure of stomata. However, in the present investigation the reduction in the rate of transpiration of the test plants, in addition to relative stomatal closure, was associated with reduced stomatal frequency. Seed presoaking with  $GA_3$  or IAA induced a significant increase in transpiration rate and stomatal frequency (Table 4 and 5).

The value of dry matter yield of the tests plants was generally lowered by increasing the salinizing agent concentration (Table 6). These inhibitory

Table 5

*Effect of seed presoaking with  $GA_3$  and IAA on leaf area ( $dm^2/plant$ ), transpiration rate ( $g/dm^2/day$ ) and stomatal frequency (number of stomata/ $mm^2$ ) of safflower plants stressed for 15 days with different levels of sodium chloride*

Treatment	Leaf area	Transpiration rate	Stomatal frequency	
			Upper epidermis	Lower epidermis
Control	0.77	2.88	183	237
- 3 bar	0.68**	2.85	158**	215**
- 6 bar	0.53**	2.60**	100**	169**
- 9 bar	0.36**	1.77**	66**	158**
- 12 bar	0.29**	1.14**	50**	83**
Control + $GA_3$	0.83*	3.31*	162*	266**
- 3 bar + $GA_3$	0.70	3.10**	169	236**
- 6 bar + $GA_3$	0.59*	2.77*	125**	225**
- 9 bar + $GA_3$	0.43*	1.77	100**	183**
- 12 bar + $GA_3$	0.39**	1.18	50	100*
Control + IAA	0.86**	3.33**	168	258**
- 3 bar + IAA	0.71	3.29**	158	258**
- 6 bar + IAA	0.59*	2.77*	133**	222**
- 9 bar + IAA	0.43*	1.79	100**	147
- 12 bar + IAA	0.37**	1.31*	50	107**
L.S.D. at 5%	0.06	0.14	16.03	13.37
L.S.D. at 1%	0.08	0.19	21.03	17.92

\* Significant differences.

\*\* Highly significant differences as compared with the control.

Table 6

*Effect of seed presoaking with GA<sub>3</sub> and IAA on the dry matter yield (g/plant) of maize and safflower plants stressed with different levels of sodium chloride (45 days in the case of maize and 60 days that of safflower)*

Treatment	Whole plant	Shoot/root ratio	Treatment	Whole plant	Shoot/root ratio
Control	2.10	0.46	Control	1.11	1.70
-2 bar	2.16	0.61**	-3 bar	0.05**	2.51**
-4 bar	1.62**	0.71**	-6 bar	0.38**	3.13**
-6 bar	0.91**	0.75**	-9 bar	0.17**	3.77**
-8 bar	0.75**	0.67**	-12 bar	0.06**	2.21*
-10 bar	0.68**	0.58**			
Control+GA <sub>3</sub>	3.44**	0.44	Control+GA <sub>3</sub>	0.91**	2.04
-2 bar+GA <sub>3</sub>	2.47**	0.72**	-3 bar+GA <sub>3</sub>	0.87	1.66**
-4 bar+GA <sub>3</sub>	1.81*	0.79*	-6 bar+GA <sub>3</sub>	0.54*	3.77*
-6 bar+GA <sub>3</sub>	1.22**	0.72	-9 bar+GA <sub>3</sub>	0.22	3.02**
-8 bar+GA <sub>3</sub>	1.11**	0.37**	-12 bar+GA <sub>3</sub>	0.10	3.30**
-10 bar+GA <sub>3</sub>	1.02**	0.38**			
Control+IAA	3.13**	0.51	Control+IAA	1.73**	2.04
-2 bar+IAA	2.73**	0.54	-3 bar+IAA	0.05	2.01
-4 bar+IAA	1.69	0.66	-6 bar+IAA	0.54*	2.38**
-6 bar+IAA	1.34**	0.84*	-9 bar+IAA	0.29	6.83**
-8 bar+IAA	0.87	0.36**	-12 bar+IAA	0.11	3.95**
-10 bar+IAA	0.06*	0.34**			
L.S.D. at 5%	0.18	0.08	L.S.D. at 5%	0.13	0.51
L.S.D. at 1%	0.24	0.10	L.S.D. at 1%	0.18	0.69

\* Significant differences.

\*\* Highly significant differences.

effects support the results obtained by some other authors using various plants (Hutton, 1971; Lashin and Atanasiu, 1972; Haikal, 1975; Nassery et al., 1979; Coughlan and Wynjones, 1980; Joshi and Naik, 1980; Singh and Singh, 1980; Ralph et al., 1984 and Nerson and Paris, 1984). It was noticed that the growth of roots was more severely retarded by salt-stress than was the growth of shoots, which could be clearly noticed from the increased values of the shoot/root ratio (Table 6). The close correspondence between the retarded growth and salinization was ascribed to the effect of NaCl on several factors of plant activities such as osmotic adjustment (Bernstein, 1963), protein and nucleic acid synthesis (Nieman, 1965; and Bejaoui, 1985); ion uptake (Greenway et al., 1966), hormonal balance (Itai et al., 1968), enzyme activities (Weimberg, 1970) and photosynthesis (Ahmed et al., 1979b). The beneficial effect of seed presoaking with GA<sub>3</sub> or IAA on the growth of salinized plants was revealed in this work (Table 6). This may be attributed to the increased uptake of water which is the consequence rather than the cause of cell expansion induced by hormonal treatments. It is possible that, under the influence



of salt-stress, the level of naturally synthesized growth hormones may be suppressed and that the exogenous application of phytohormones supplies more or less sufficient quantities, which are implicated in growth promotion. This opinion was favoured by Shah and Loomis (1965); Hsiao (1973); Bernstein (1975); and Boucaud and Ungar (1976).

### References

- Adams, F., Bingham, T., Kaufmann, M. R., Yermanos, D. M. (1978): Responses of stomata and water, osmotic and turgor potentials of Jojoba to water and salt stress. *Agron J.*, **70**, 381-387.
- Ahmed, A. M., Heikal, M. M., Shaddad, M. A. (1979a): Changes in some plant water relationship parameters of some oil producing plants over a range of salinity stresses. *Biologia Plantarum (Praha)*, **21**, 259-265.
- Ahmed, A. M., Heikal, M. M., Shaddad, M. A. (1979b): Growth, photosynthesis and fat content of some oil producing plants influenced by some salinization treatments. *Phyton*, **19**, 259-267.
- Ahmed, A. M., Heikal, M. M., Shaddad, M. A. (1980a): Effects of salinization treatments on growth and some related physiological activities of some leguminous plants. *Cand. J. Pl. Sci.*, **60**, 713-720.
- Ahmed, A. M., Heikal, M. M., Shaddad, M. A. (1980b): Changes in growth, photosynthesis and fat content of some oil producing plants over a range of salinity stress. *Acta Agonomica* (in press).
- Badour, S. S. A. (1959): *Analytisch chemische Untersuchung des Kaliummangels bei Chlorella im Vergleich mit anderen Mangelzuständen*. Ph. D. Dissertation Goettingen.
- Bejaoui, M. (1985): Interactions between NaCl and some phytohormones on soybean growth. *J. Plant Physiol.* **120**, 95-110.
- Bernstein, L. (1963): Osmotic adjustment of plants to saline media. 11. Dynamic phase. *Am. J. Bot.*, **48**, 909-918.
- Bernstein, L. (1975): Effects of salinity and sodicity on plant growth. *Ann. Rev. Phytopathology*, **13**, 295-312.
- Boucaud, J., Unger, I. A. (1976): Influence of hormonal treatments on the growth of two halophytic species of Suaeda. *Am. J. Bot.*, **63** (5), 695-699.
- Bozcuk, S. (1975): *Effect of sodium chloride upon growth and transpiration in Statice sp. and Pisum sativum L.* Proc. of the third Mpp Meeting IZMIR. 37-42.
- Chhipa, B. R., Lal, P. (1978): Effect of pre-soaking of seeds with salt and hormone solutions and different quality waters on wheat. *J. Indian Soc. Soil. Sci.*, **26** (4), 390-396.
- Coughlan, S. I., Wynjones, S. J. (1980): Some response of *Spinacea oleracea* to salt stress. *J. Exp. Bot.*, **123**, 983-993.
- Darra, B. L., Seth, S. P., Singh, H., Mendirattas, R. S. (1973): Effect of hormone-directed presoaking on emergence and growth of osmotically stressed wheat (*Triticum sativum* L.) seed. *Agron. J.* **65**, 292-295.
- Eshel, A. (1985): Response of *Suaeda aegyptiaca* to potassium chloride, sodium chloride and sodium sulfate treatments. *Physiol Plant.* **64**, 308-315.
- Fales, F. W. (1951): The assimilation and degradation of carbohydrates by yeast cells. *J. Biol. Chem.*, 193-213.
- Gale, J., Kohl, H. C., Hagan, R. M. (1967): Changes in the water balance and photosynthesis of onion, bean and cotton plants under saline conditions. *Physiol. Plant.*, **20**, 408-420.
- Gary, O. K., Srivastava, M. P. (1970): *Yield response of I. R. B. to presowing chemical treatment*. Proc. 58th session of Indian Science Congress. Part 3, No. 86.
- Greenway, H., Gunn, A., Thomas, D. A. (1966): Plant response to saline stress rates. VIII. Regulation of ion concentration in salt-sensitive and halophytic species. *Aust. J. Biol. Sci.*, **19**, 741-756.
- Heikal, M. M. D. (1975): Physiological studies on salinity. 1. Effect of saline irrigation on growth and photosynthesis pigments of safflower and sunflower plants. *Bull. Fac. Sci., Assiut University*, **4** (1), 1-11.
- Heikal, M. M. D., Ahmed, A. M., Shaddad, M. A. (1980): Changes in dry weight and mineral composition of some oil producing plants over a range of salinity stresses. *Biologia Plantarum (Praha)*, **22** (1), 25-33.



- Heikal, M. M., Ahmed, A. M., Ismail, A. (1981b): Effect of salinization treatments on growth, pigment contents and mineral composition of *Datura stramonium* Bull. Fac. Sci., Assiut University (in press).
- Heikal, M. M. D., Shaddad, M. A., Ahmed, A. M. (1982a): Effect of water stress and gibberellic acid on germination of flax, sesame and onion seeds. *Biologia Plantarum (Praha)*, **24** (2), 124-129.
- Hsiao, T. C. (1973): Plant response to water stress. *Ann. Rev. Plant Physiol.* **24**, 519-570.
- Hutton, E. M. (1971): Variation in salt response between tropical pasture legumes. *SABRAO New Sletter* **3**, 75-81.
- Lkuma, H., Thimann, K. V. (1963): The role of seed coat in germination of photosensitive lettuce seeds. *Plant Cell Physiol.*, **4**, 169-185.
- Itai, C., Richmond, A., Vaadia, Y. (1968): The role of root cytokinins during water and salinity stress. *Israel J. Bot.*, **17**, 187-195.
- Jeffrey, R., Critchley, C. (1985): Effects of salt stress on the growth, ion content, stomatal behaviour and photosynthetic capacity of a salt-sensitive species, *Phaseolus vulgaris*. *Planta (Berl.)* **164**, 151-162.
- Jensen, C. R. (1975): Effect of salinity in the root-medium. 11. Photosynthesis and transpiration in relation to superimposed water stress from change of evaporative demands and of root temperature. *Acta Agriculturae Scandinavica*, **25**, 72-80.
- Joshi, G. V., Naik, G. R. (1980): Response of sugarcane to different types of salt stress. *Plant and Soil*, **56**, 255-263.
- Kaufmann, M. R., Ross, K. J. (1970): Water potential, temperature and kinetic effects on seed germination in soil and solute systems. *Amer. J. Bot.*, **57**, 413-419.
- Kessler, B. (1961): Nucleic acid as factors in drought resistance in higher plants. *Rec. Advance, Bot.*, **2**, 1153-1159.
- Khan, A. H., Naqvi, S. S. M. (1984): The effect of sodium chlorid and polyethylene glycol on germination and water contents of 2 mung bean (*Phaseolus aureus*) varieties. (Plant physiol. Div., Atomic Energy. Agric. Res. Cont., Tandojam, Pakistan.) *PAK, J. Bot.* **16**, 123-128.
- Lashin, M. H., Atanasiu (1972): Studies on the effect of salt concentration formation of dry matter, uptake of mineral nutrients and mineral composition of cotton plants during the vegetative growth period. 2. *Acker and Pflanzenbau*, **135**, 178-186.
- Lin, J. Y. (1985): Effect of plant growth regulators on seed germination and seedling growth of maize under saline stress. *J. of Agric. Association of China*, **131**, 1-9.
- Maftoun, M., Sepaskhah, A. R. (1978): Effect of temperature and osmotic potential on germination of sunflower and safflower and on hormone-treated sunflower seeds. *Can. J. Plant. Sci.* **58**, 295-301.
- Meiri, A., Poljakoff-Mayber, A. (1970): Effect of various salinity regimes on growth, leaf expansion and transpiration rate of bean plants. *Soil. Sci.* 26-34.
- Melzack, R. N., Bravdo, B., Riov, J. (1985): The effect of water stress on photosynthesis and related parameters in *Pinus halepensis*. *Phasiol. Plant.* **64**, 294-300.
- Nassery, H., Ogata, G., Mans, E. V. (1979): Sensitivity of sesame to various salts. *Agron. J.*, **71**, 595-597.
- Nerson, H., Paris, H. S. (1984): Effects of salinity on germination seedling growth and yield of melons. *Irrig. Sci.*, **5**, 265-274.
- Nieman, R. H., Bernstein, L. (1959): Interactive effects of gibberellic acid and salinity on the growth of beans. *Amer. J. Bot.*, **46** (9), 667-670.
- Nieman, R. H. (1965): Expansion of bean leaves and its suppression by salinity. *Plant. Physiol.*, **40**, 156-161.
- Paech, K., Tracey, M. V. (1956): *Modern Methods of Plant Analysis. Vol. I.* Springer-Verlag, Berlin.
- Parmer, M. T., Moore, R. P. (1968): Carbowax 6000, mannitol and sodium chloride for simulating drought conditions: germination studies of corn (*Zea mays* L.) of strong and weak vigor. *Agron. J.*, **60**, 192-195.
- Patil, P. K., Patil, V. K., Ghonsikar, C. P. (1984): Effect of soil salinity on growth and nutritional status of guava (*Psidium guajava*). *Int. J. Trop. Agric.*, **2**, 337-344.
- Poljakoff-Mayber, A., Gale, J. (1975): *Ecological studies. 15. Plants in saline environments* (pp. 197). Springer-Verlag, Berlin.
- Ralph, W. K., Emanuel, E., Robert, W. P. (1984): Physiological responses to salinity in selected lines of wheat plant. *Plant Physiol.*, **74**, 417-423.
- Schlegel, H. G. (1956): Die Verwertung organischer Sauren durch *Chlorella* in Licht. *Planta*, **47**, 510.
- Shaddad, M. A., Heikal, M. M. (1981): Effect of salinization treatments on growth, fat content



- and some plant-water relation parameters of cotton, safflower, and lupine plants. *Bull. Fac. Sci., Assiut Univ.* (in press).
- Shaddad, M. A., Heikal, M. M. (1982): Interactive effect of gibberellic acid and salinity on Kidney bean. *Bull. Fac. Sci. Assiut Univ., Egypt*, **11**, 135-149.
- Shah, C. B., Loomis, R. S. (1965): Ribonucleic acid and protein metabolism in sugarbeets during drought. *Physiol. Plant.*, **18**, 240-254.
- Singh, G., Singh, J. (1980): Effect of growth regulation on the growth parameters of chick-pea (*Cicer arietinum*) growth under different salinity levels. *Indian J. Agric. Sci.*, **50** (1), 23-30.
- Sionit, N., Kheradnam, M., Ghorashy, S. R. (1973): Effect of different osmotic potentials of media on germination of three safflower varieties. *Physiol. Plant.*, **29**, 272-273.
- Stewart, F. C. (1959): *Plant physiology, a treatise analysis of growth regulating substances*. Academic Press, New York, 131-181.
- Stewart, J. I., Hagan, R. M., Pruitt, W. O. (1976): *Salinity effects on corn yield, evapotranspiration, leaching fraction and irrigation*. Proceeding of the International Conference on managing saline water for irrigation planting for the fruit (Dregne, H. E. Editor) 316-332.
- Stout, D. G., Simpson, G. M., Flotre, D. M. (1980): Drought resistance of *Sorghum bicolor* L. Moench. 3. Seed germination under osmotic stress. *Can. J. Plant. Sci.*, **60**, 13-14.
- Tal, M., Gavish, U. (1973): Salt tolerance in the wild and the cultivated tomato, water balance and abscisic acid in *Lycopersicum esculentum* L. and *Peruvianum* under low and high salinity, *Aust. J. Agric. Res.*, **24**, 253-361.
- Verasan, V., Phillips, R. E. (1978): Effect of soil water stress on growth and nutrient accumulation of corn. *Agron. J.*, **70**, 613-619.
- Waisel, Y. (1972): *Biology of halophytes*. New York, Acad. Press.
- Weimberg, R. (1970): Enzyme levels in pea seedling grown on highly salinized media. *Plant. Physiol.*, **46**, 466-470.
- Wong, C. H., Jager, H. T. (1978): Salt induced vasculature in mesophyll cells of *Atriplex* species. *Plant Science Letters*. **12**, 63-68.
- Zidan, M. A. (1979): *Photosynthetic activity, mineral composition and growth of some legumes as affected by salinity*. M. Sc. Thesis, Assiut University, Assiut Egypt.

## SALINITY-HORMONE INTERACTION IN RELATION TO THE CHEMICAL COMPOSITION OF MAIZE AND SAFFLOWER PLANTS

A. F. RADI,\* M. M. HEIKAL,\* A. M. ABDEL-RAHMAN\*\*  
and B. A. A. EL-DEEP\*\*

\* DEPARTMENT OF BOTANY, FACULTY OF SCIENCE ASSIUT UNIVERSITY, ASSIUT, EGYPT

\*\* DEPARTMENT OF BOTANY, FACULTY OF SCIENCE. ASSIUT UNIVERSITY, SOHAG, EGYPT

(Received 10 July 1987; accepted 23 November 1987)

The interactive effect of salinity and growth hormones on the chemical composition of maize and safflower plants was investigated.

The photosynthetic pigment contents of maize plants increased with the rise of NaCl salinization and the opposite trend was exhibited by safflower plants. Seed presoaking with GA<sub>3</sub> or IAA generally induced a significant increase in the values of the total pigment contents of the test plants.

The mineral composition of the test plants showed somewhat variable responses to salinization and hormone treatments.

The production of soluble, insoluble as well as total carbohydrates by maize plants were reduced with the rise of salinization level. On the other hand, the pattern of changes in carbohydrate content of safflower plants was irregular for the different salinity and hormone treatments.

**Keywords:** *Carthamus tinctorius* L., mineral composition, plant hormones, salinity, *Zea mays* L.

### Introduction

The estimation of the total photosynthetic pigment contents and the different pigment fractions has been adopted in various studies, particularly in relation to salt-stress. Many investigators found that salinization exerts a general reduction in the contents of the photosynthetic pigments (Ashour and Thalooth, 1971; Shimose, 1973; Heikal, 1975; and Ahmed et al., 1980). On the other hand, Dostanova (1966) found that the leaves of salinized sugar beet plants are more rich in chlorophylls and carotenoids, compared with those grown under non-salinized conditions.

As regards nitrogen metabolism, salinized plants exhibited some disturbances in nitrogen metabolism (Wilson et al., 1970; El-Shourbagy and Missak, 1975; Patil et al., 1984; and Fayez, 1985). In this respect, the protein contents of various plant tissues were found to decline under saline conditions.

Salinity stress seems to affect plant carbohydrate metabolism in several ways (Todd and Basler, 1965). Earlier studies indicated that carbohydrate contents were frequently higher in plants grown under saline conditions,



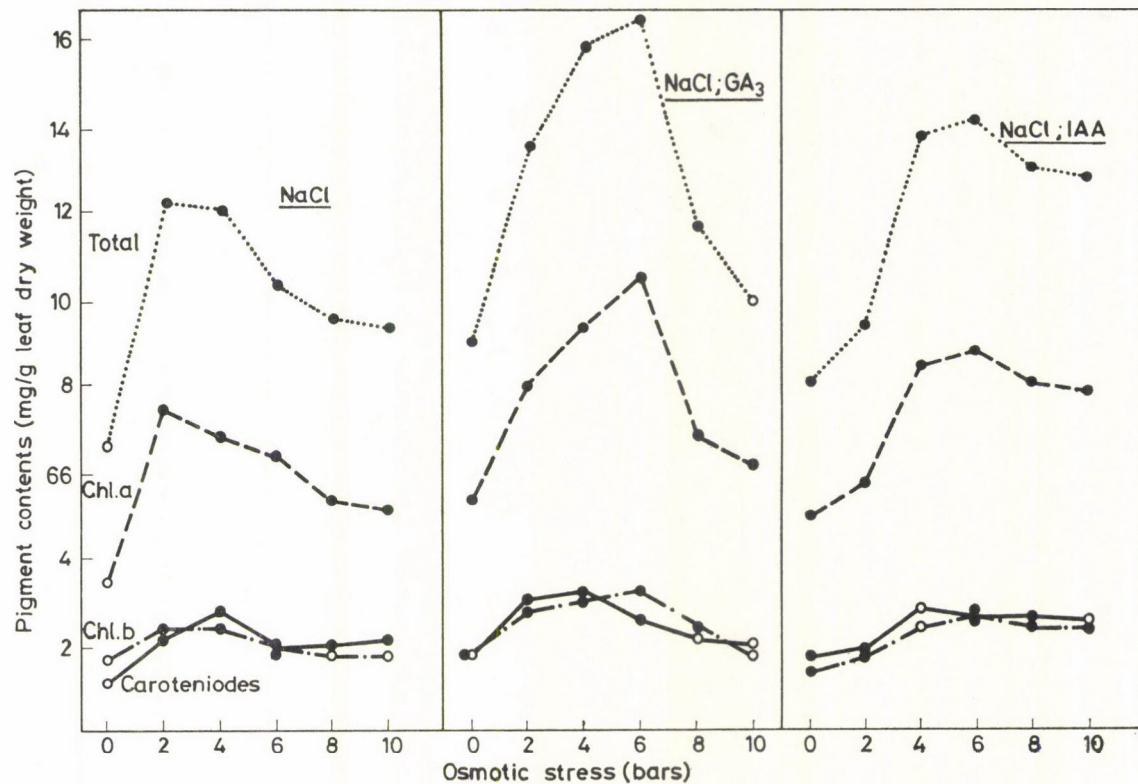


Fig. 1. Effect of seed presoaking with phytohormones on pigment contents of leaves of maize plants stressed for 45 days with different levels of sodium chloride. Black circles indicate highly significant differences, half-filled circles indicate significant differences, empty circles = no significant differences. ○ = control treatment

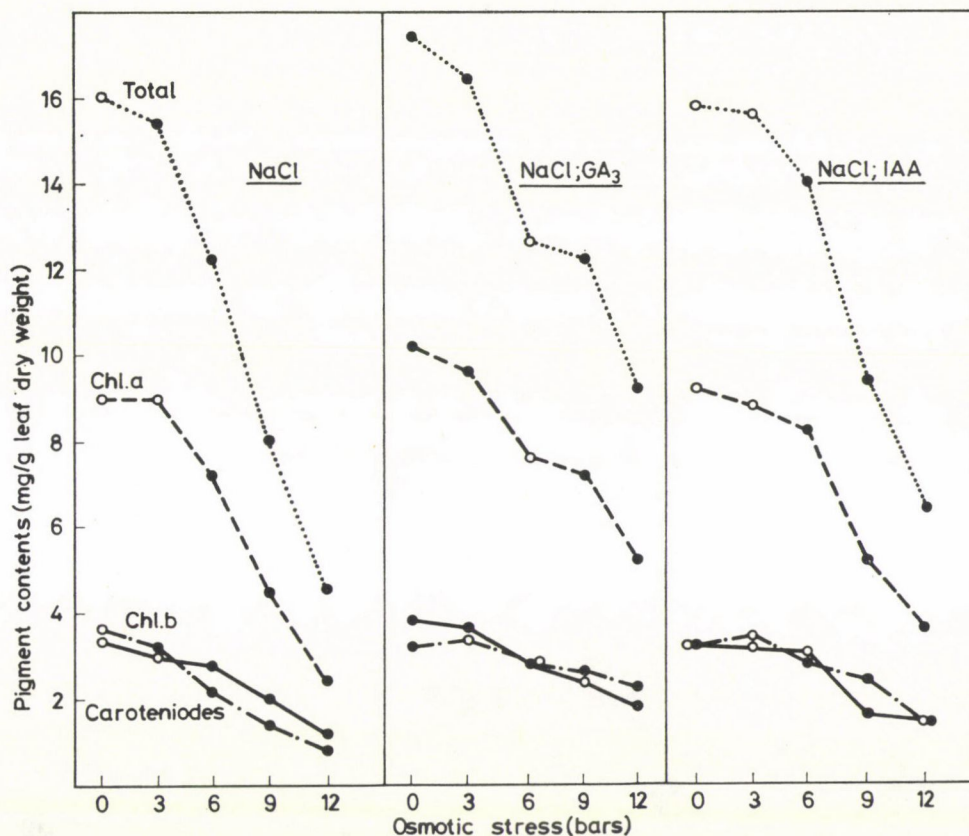


Fig. 2. Effect of seed presoaking with phytohormones on pigment contents of leaves of safflower plants stressed for 60 days with different levels of sodium chloride. Black circles indicate highly significant differences, half-filled circles indicate significant differences empty circles = no significant differences. ○ = control treatment

although exceptions were noted (Bernstein and Hayward, 1958; Barlow et al., 1976; El-Shahaby, 1981 and Fayez, 1985).

With respect to ion uptake, a number of investigators demonstrated that nutrient uptake by certain plant species is curtailed by salinization (Shimose, 1968; Lashin and Atanasiu, 1972; El-Shourbagy and Missak, 1975; Patil et al., 1984 and Eshel, 1985). Other investigators showed that salinization treatments resulted in a promotion rather than an inhibition of nutrient uptake (Shimose, 1963 and 1964; Asana and Kale, 1965; Mehrotra, 1971 and Heikal et al., 1980a).

Presoaking seeds with optimal concentration of certain phytohormones has been proven beneficial for the growth and yield of some crop species living under saline conditions, by increasing nutrient reserves through increased



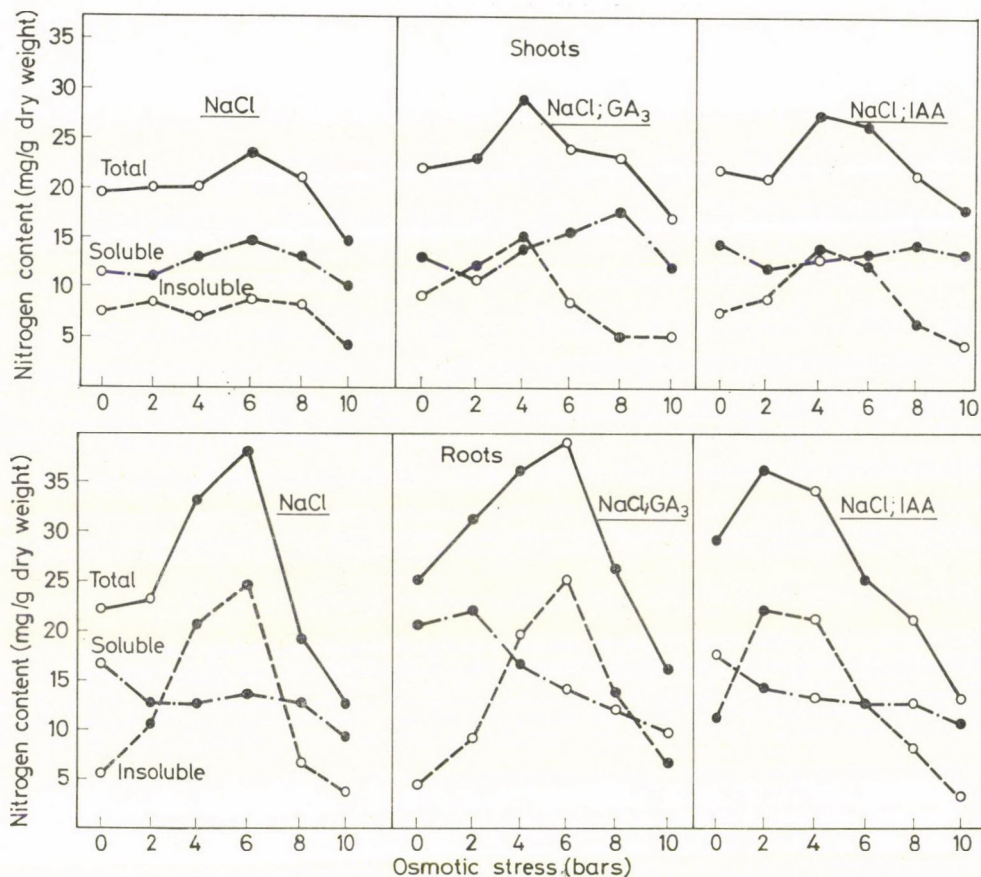


Fig. 3. Effect of seed presoaking with phytohormones on nitrogen content in shoots and roots of maize plants stressed for 45 days with different levels of sodium chloride. Black circles indicate highly significant differences, half-filled circles indicate significant differences, empty circles = no significant differences.  $\circ$  = control treatment

physiological activities and root proliferation (Darra et al., 1973; Chippa and Lal, 1978; and Shaddad and Heikal, 1982).

The present work was undertaken to study the effect of various concentrations of NaCl on the chemical composition of maize (*Zea mays*) and safflower (*Carthamus tinctorius*) plants. The role of GA<sub>3</sub> and IAA in modifying the salt-stress induced changes was also investigated.

### Materials and methods

The optimum conditions reached from preliminary tests conducted to determine the optimum hormone concentration, as well as the optimum period of presoaking and rying, were exactly the same as described in a previous investigation (in press). Seeds of test plants were

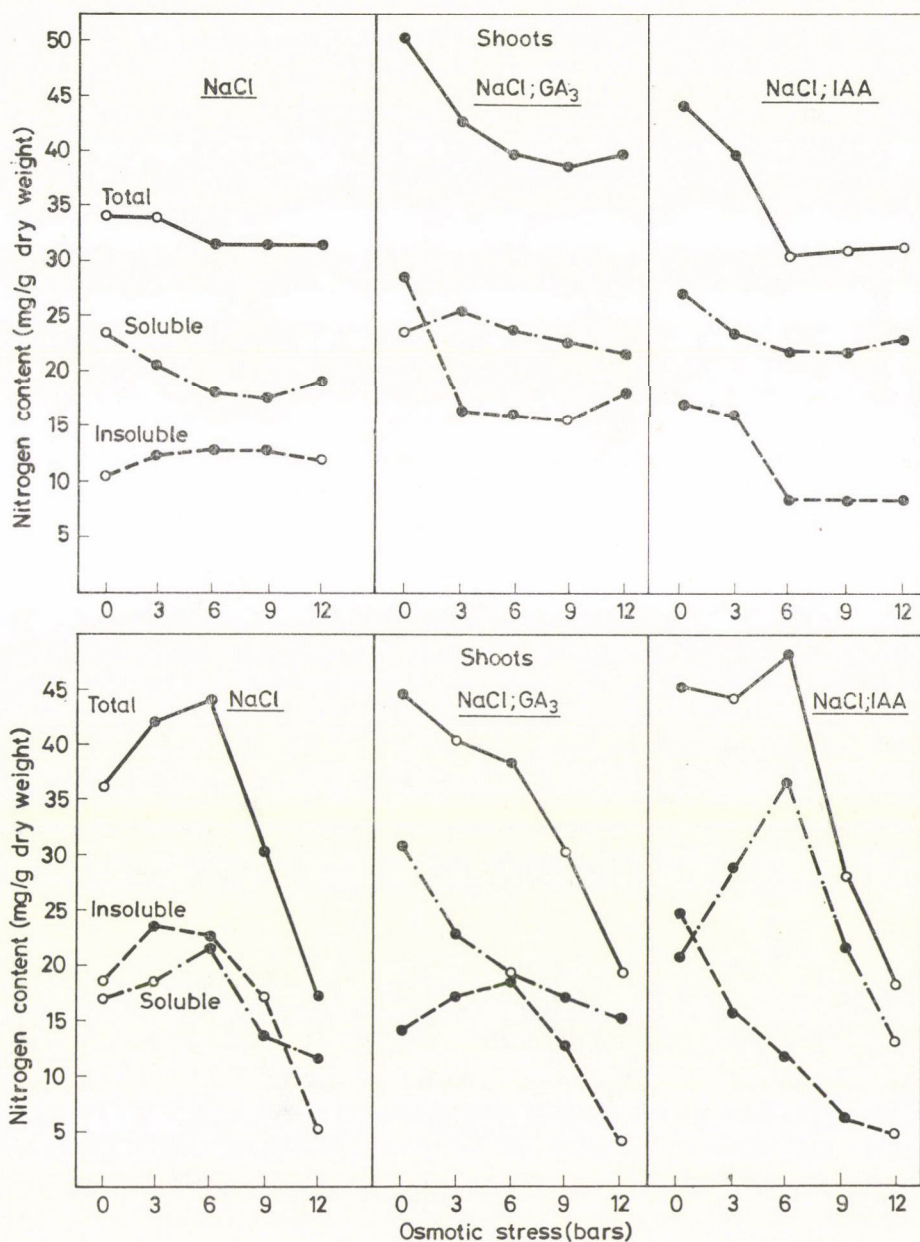


Fig. 4. Effect of seed presoaking with phytohormones on nitrogen content in shoots and roots of safflower plants stressed for 60 days with different levels of sodium chloride. Black circles indicate highly significant differences, half-filled circles indicate significant differences, empty circles = no significant differences. O = control treatment



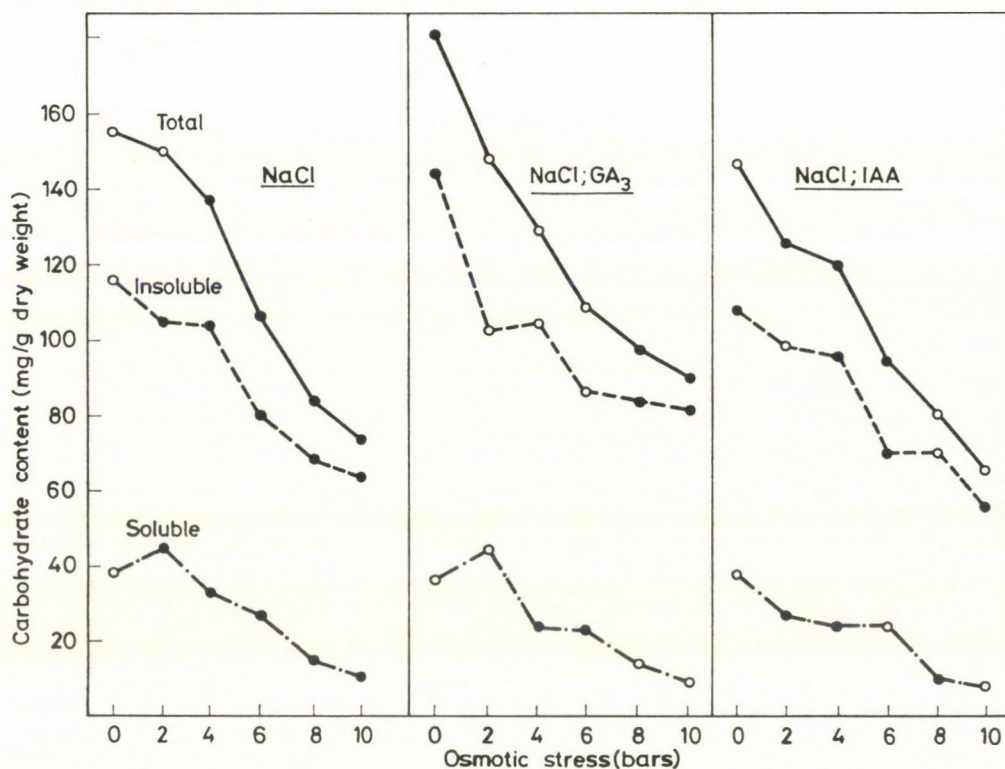


Fig. 5. Effect of seed presoaking with phytohormones on carbohydrate content in the whole plant body of maize plants stressed for 45 days with different levels of sodium chloride. Black circles indicate highly significant differences, half-filled circles indicate significant differences, empty circles = no significant differences.  $\circ$  = control treatment

presoaked in  $GA_3$  or IAA and after being dried were sown in plastic pots containing 2 kg air-dried soil (sand/clay 1 : 1 v/v). Using NaCl and N/10 Pfeffer's nutrient solution, stress levels were chosen at -2, -4, -6, -8 and -10 bar in the case of maize plants and -3, -6, -9 and -12 bar in that of safflower plants. The test plants were irrigated with an experimental solution every other day for two weeks from the beginning of planting and the soil moisture content was kept near the field capacity. Thereafter, the salinized and non-salinized plants were irrigated every other day with N/10 Pfeffer's solution. The plants were left to grow under the various treatments in a greenhouse for 45 days in the case of maize and 60 days in that of safflower.

The photosynthetic pigments (chlorophyll-a, chlorophyll-b and carotenoids) were determined using the spectrophotometric method recommended by Metzner et al. (1965).

Nitrogen was determined by the micro-Kjeldahl method (Paech and Tracey, 1956).

The anthrone sulphuric acid method (Fales, 1951; Schlegel, 1956 and Badour, 1959) was used for the determination of carbohydrates.

Dry samples were ground into a fine powder in a micromill and assayed for mineral ion determination. The "flame photometric" method was used for both sodium and potassium determinations (Williams and Twine, 1960). The versene (disodium dihydrogen ethylenediamine tetra acetic acid) titration method (Schwarzenbach and Biederman, 1948) was employed for both calcium and magnesium determinations.

The data were statistically analysed and the least significant differences were obtained (Snedecor and Cochran, 1967).

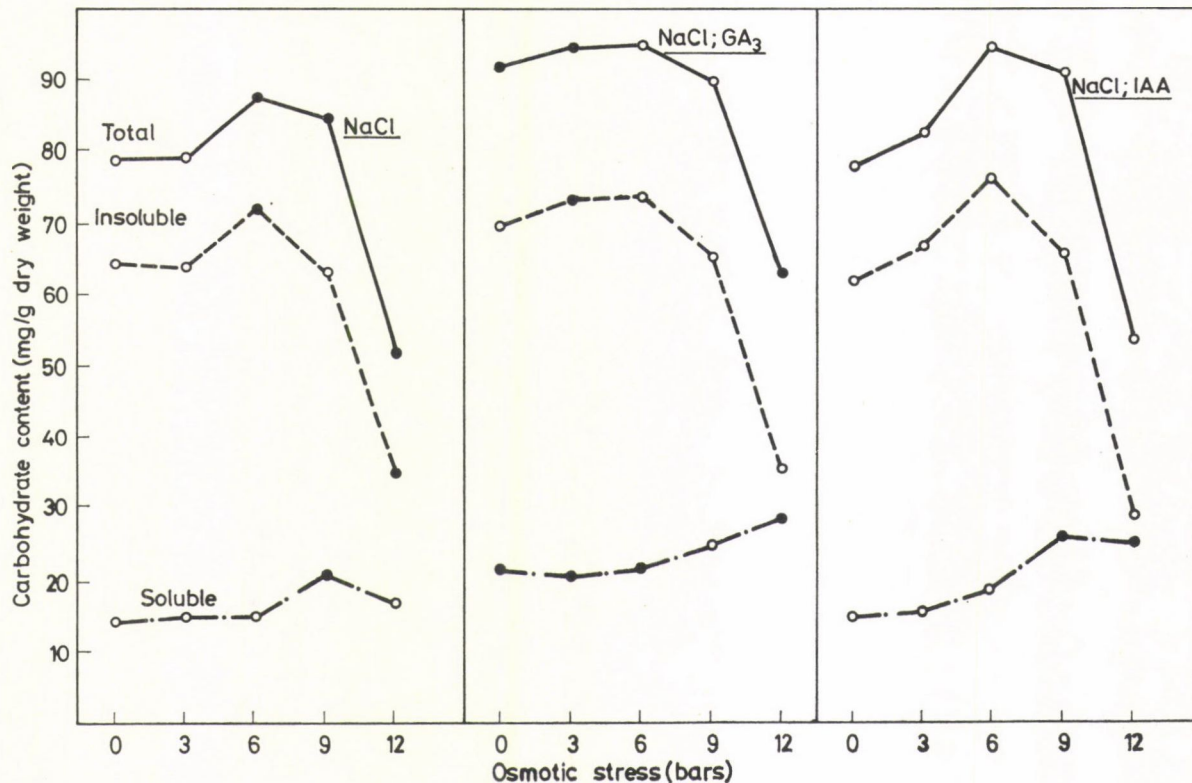


Fig. 6. Effect of seed presoaking with phytohormones on carbohydrate content in the whole plant body of safflower plants for 60 days with different levels of sodium chloride. Black circles indicate highly significant differences, half-filled circles indicate significant difference, empty circles = no-significant differences. ○ = control treatment



## Results and discussion

The biosynthesis of pigments in salinized maize plants was markedly activated by salt-stress (Fig. 1). These results are generally in accordance with those obtained by Ahmed et al. (1977, 1978) and Heikal et al. (1980c). In the case of safflower plants, the production of pigments was generally reduced (Fig. 2). Similar results were also obtained by some other authors using different kinds of plants (Ashour and Thalooh, 1971; Ahmed et al., 1979; Joshi and Naik, 1980 and Chavan and Karadge, 1980). Seed presoaking with  $GA_3$  or IAA induced a significant increase in the values of the total pigment contents of the test plants (Figs 1 and 2). This promotive effect of seed presoaking obtained in this experiment agrees with the results obtained by Varshney and Baijal (1979); Shaddad and Heikal (1982) and El-Tayeb (1986), using some grasses and kidney bean plants.

The nitrogen fractions of the test plants exhibited variable values (Figs 3 and 4). These variable changes support the results obtained by Wilson et al. (1970); El-Shourbagy and Missak (1975) and Heikal (1976), who demonstrated that salinity induces gross disturbances in the nitrogen metabolism of certain plants. However, in this present investigation, regardless of salinity level, seed presoaking with  $GA_3$  induced a significant increase in soluble, insoluble, and consequently, the total nitrogen in safflower plants (Fig. 3).

This response would suggest that the deteriorative effect of salt on plant growth and other relevant physiological activities can either be alleviated or modified to some extent by treating the seeds with the proper concentrations of a suitable growth promoter.

Salt-stress was found to induce a significant reduction in the carbohydrate contents of maize plants (Fig. 5). In this respect, Cecil and Stewart (1971), Barlow et al. (1976) and Ismail (1982) reported that salinity generally hydrate contents of maize plants (Fig. 5). In this respect, Cecil and Stewart (1971), Barlow et al. (1976) and Ismail (1982) reported that salinity generally affects the carbohydrate metabolism. On the other hand, the increase in carbohydrate contents of safflower plants (Fig. 6), especially at the relatively low and moderate salinization levels, could be ascribed to the increased soluble carbohydrate fractions. These results agree with those obtained by Salama et al. (1980) and Ismail (1982) using some glycophytic plants, and a mixture of NaCl and other salts as salinizing agents. Seed presoaking with the phytohormones induced an irregular effect on the production of carbohydrates in salt-stressed plants (Figs 5 and 6).

Salinization treatments were found to induce some considerable changes in the concentration of mono ( $Na^+$ ,  $K^+$ ) and divalent ( $Ca^{2+}$ ,  $Mg^{2+}$ ) cations in the main parts of the test plants. However, the magnitude and the patterns of these changes differed according to the level of salinization, the nutritive

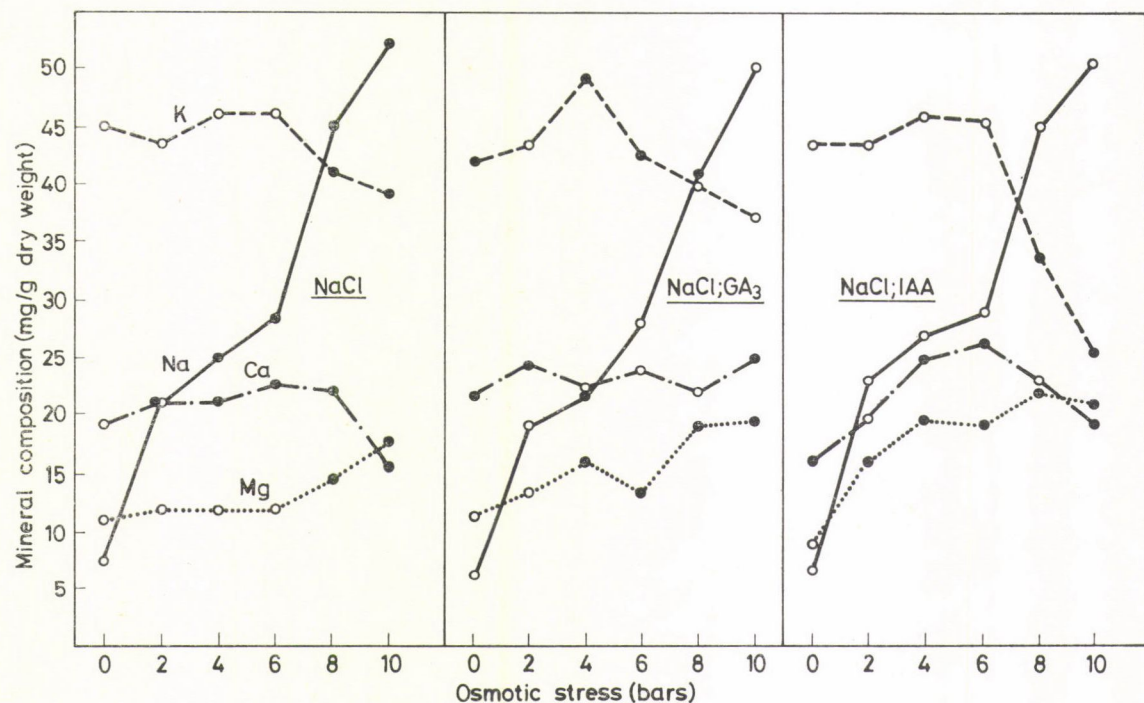


Fig. 7. Effect of seed presoaking with phytohormones on mineral composition of shoots of maize plants stressed for 45 days with different levels of sodium chloride. Black circles indicate highly significant differences, half-filled circles indicates significant differences, empty circles = no significant differences. ○ = control treatment



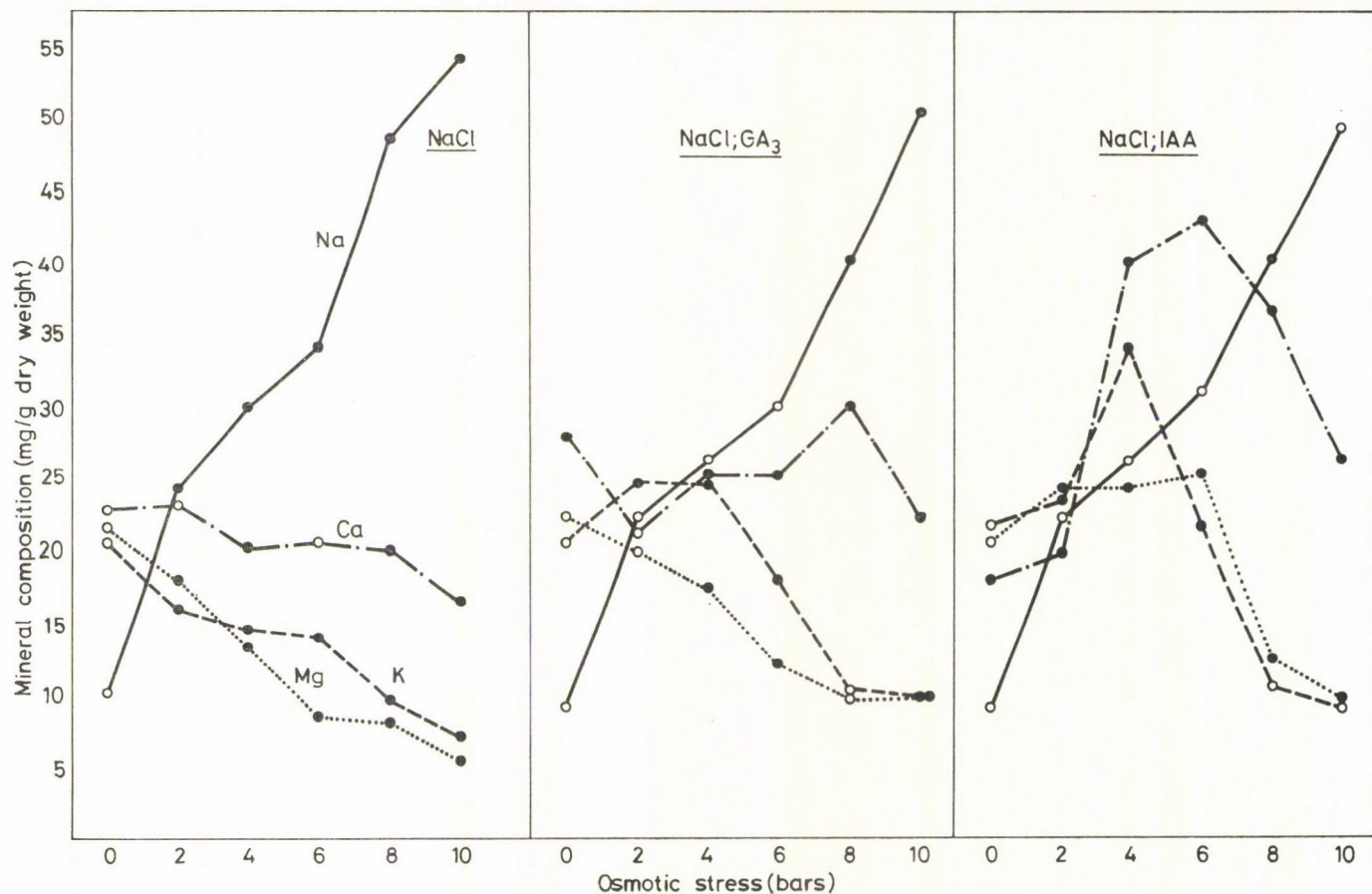


Fig. 8. Effect of seed presoaking with phytohormones on mineral composition of roots of maize plants stressed for 45 days with different levels of sodium chloride. Black circles indicate highly significant differences, half-filled circles indicate significant differences, empty circles = no significant differences.  $\circ$  = control treatment

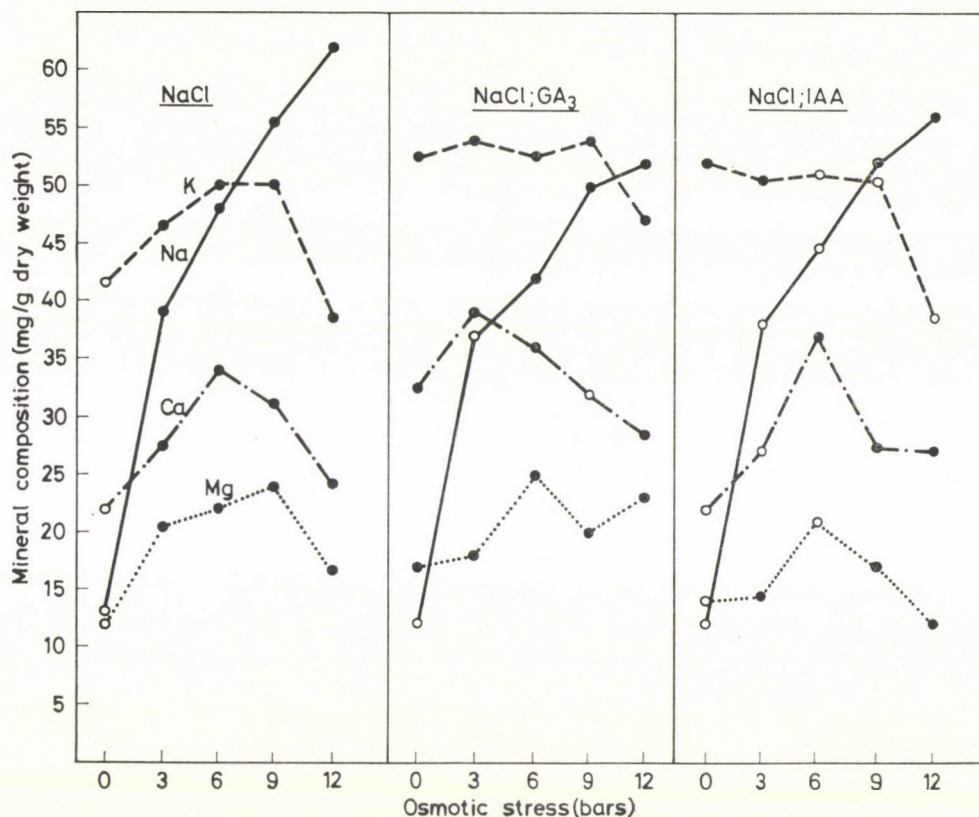


Fig. 9. Effect of seed presoaking with phytohormones on mineral composition of shoots of safflower plants stressed for 60 days with different levels of sodium chloride. Black circles indicate highly significant differences, half-filled circles indicate significant differences, empty circles = no significant differences.  $\circ$  = control treatment

element, the plant type and the organ harvested (Figs 7, 8, 9 and 10). The increase in sodium concentration, with the rise of salinization level, accords with the results obtained by other investigators, using some other crop plants (Yousif et al., 1972; Heikal et al., 1979 and Coughlan and Wynjones, 1980). However, the extent of sodium accumulation varied among the main plant parts of the test plants. In this respect, the shoots accumulated generally lower amounts of sodium than the roots. This is in accordance with Jacoby (1965); Lahaye and Epstein (1969) and Heikal et al. (1980b).

The reduction of potassium concentration in salinized maize plants conforms with the results obtained by Rush and Epstein (1976); Chavan and Karadge (1980) and Heikal et al. (1981). In safflower plants, however, relatively low and moderate levels of NaCl promoted the accumulation of potassium



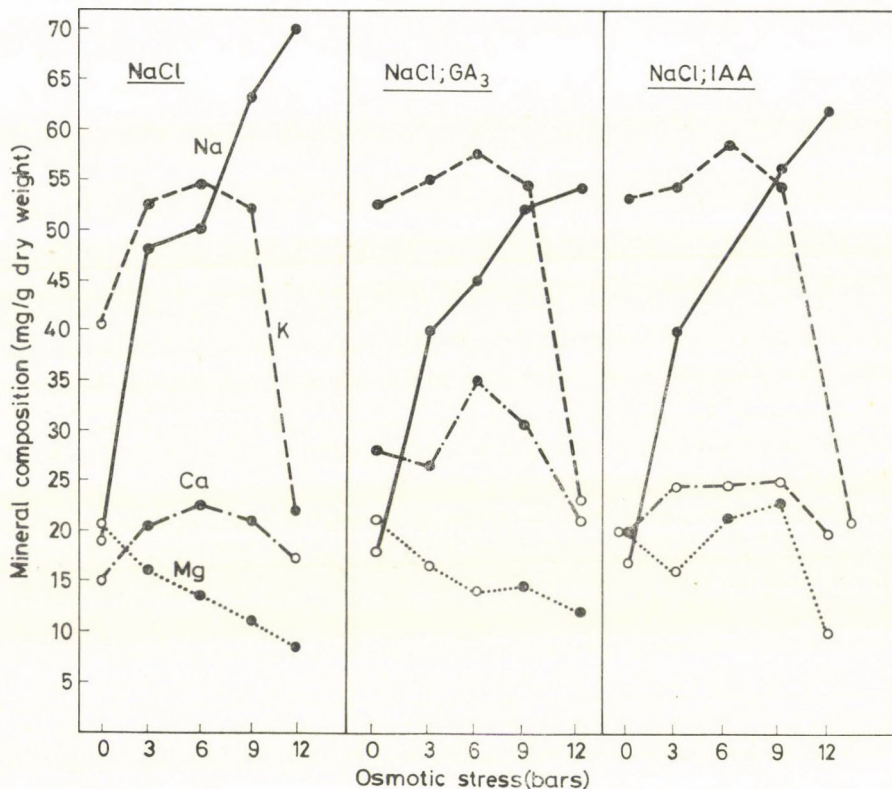


Fig. 10. Effect of seed presoaking with phytohormones on mineral composition of roots of safflower plants stressed for 60 days with different levels of sodium chloride. Black circles indicate highly significant differences, half-filled circles indicate significant differences, empty circles = no significant differences. ○ = control treatment

(Fig. 9 and 10). This trend was also described by Wilson et al. (1970) and Heikal et al. (1980a and b).

The concentration of calcium in the shoots and roots of safflower (Figs 9 and 10) increased with salinity; while in maize, calcium concentration generally exhibited irregular trends (Figs 7 and 8).

Magnesium concentration in the main parts of the salt-affected plants showed variable alterations (Figs 7, 8, 9 and 10). This trend was also described by authors who used other plants (Bierhuizen and Ploegman, 1967 and Helal and Mengal, 1979).

The alteration in distribution and accumulation of mono and divalent cations in the different organs of salt-stressed plants may be one aspect of the role of these cations in regulating the physiological activities of these plants. Evidence to support this suggestion may be obtained from the work of Radi (1964), who studied the effect of other unfavourable environmental

factors on the absorption and distribution of mineral elements in maize, sunflower and tomato plants. He concluded that, under unfavourable conditions, the cations are distributed and accumulated in the different plant organs in such a way that they help to regulate the functional state of these organs.

On the other hand, seed presoaking with the tested phytohormones was accompanied by an impairment of sodium accumulation in the shoots and roots.  $GA_3$  or IAA was found to induce a promotion rather than an inhibition in the accumulations of potassium, calcium and magnesium. This response strongly suggests that exogenously applied  $GA_3$  or IAA may be involved in the maintenance of their levels, to enhance the growth activities involving metabolism processes in which these nutrients are utilized.

### References

- Ahmed, A. M., Heikal, M. M., Shaddad, M. A. (1977): Photosynthesis of some economic plants as affected by salinization treatments. II. Safflower and maize. *Egypt. J. Bot.*, **20** (1), 17-27.
- Ahmed, A. M., Heikal, M. M., Shaddad, M. A. (1978): *Photosynthetic activity, pigment content and growth of Ricinus communis plant as influenced by salinization treatments*. Proc. Ann. Meeting of Saudi Arabian Bio. Soc. (in Press).
- Ahmed, A. M., Heikal, M. M., Shaddad, M. A. (1979): Growth photosynthesis and fat content of some oil producing plants influenced by some salinization treatments. *Phyton*, **19**, 259-267.
- Ahmed, A. M., Heikal, M. M., Shaddad, M. A. (1980): Effect of salinization treatment on growth and some related physiological activities of some leguminous plants. *Can. J. Plant Sci.*, **60**, 713-720.
- Asana, R. D., Kale, V. R. (1965): A study of salt tolerance of four varieties of wheat. *Indian J. Plant Physiol.*, **8**, 6-20.
- Ashour, N. I., Thalooh, A. T. (1971): Effect of saline irrigation on photosynthetic apparatus and yield of sugar best plants. *U. A. R. J. Bot.*, **14**, 221-231.
- Badour, S. S. A. (1959): *Analytisch-chemische Untersuchung des Kaliummangels bei Chlorella in Vergleich mit anderen Mangelzuständen*. Ph. D. Dissertation Goettingen.
- Berlow, E. W. R., Boersma, L., Young, J. L. (1976): Root temperature and soil water potential effect on growth and soluble carbohydrates concentration of corn seedlings. *Crop. Sci.*, **16**, 59-62.
- Bierhuizen, J. F., Ploegman, C. (1967): Salt tolerance of tomato. *Meded. Dir. Tuninb.*, **30**, 302-310.
- Chavan, P., Karadge, B. A. (1980): Influence of salinity on mineral nutrition of peanut (*Arachis hypogaea*). *Plant and Soil*, **54**, 5-13.
- Chhipa, B. R., Lal, P. (1978): Effect of presoaking of seeds with salt and hormone solutions and different quality waters on wheat. *J. Indian Soc. Soil Sci.*, **26** (4), 390-396.
- Coughlan, S. J., Wynjones, S. J. (1980): Some responses of *Spinacea oleracea* to salt stress. *J. Exp. Bot.*, **123**, 883-893.
- Darra, B. L., Seth, S. P., Singh, H., Mendiratta, R. (1973): Effect of hormone-directed presoaking on emergence and growth of osmotically stressed wheat (*Triticum sativum* L.) seeds. *Agron. J.*, **65**, 292-295.
- Dostanova, R. K. H. (1966): Effect of  $Na_2SO_4$  and NaCl on metabolism of plastid pigments in plants. *F. Biologia Rast.*, **13**, 614-622.
- El-Shahaby, O. A. (1981): *Studies on growth and metabolism of certain plants*. Ph. D. Thesis, University of Mansoura, Egypt.
- El-Shourbagy, M. N., Missak, N. L. (1975): Effect of growing season and salinity on growth, mineral composition and seed-lipid characteristics of some *Ricinus communis* L. varieties. *Flora*, **164**, 51-71.



- El-Tayeb, A. M. (1986): *Physiological response of some plants to the interactive effect of drought and growth regulators*. M. Sc. Thesis. Assiut Univ. Egypt.
- Eshel, A. (1985): Response of *Suaeda aegyptiaca* to potassium chloride, sodium chloride and sodium sulfate treatments. *Physiol. Plant.*, **64**, 308-315.
- Fales, F. W. (1951): The assimilation and degradation of carbohydrates by yeast cells. *J. Biol. Chem.*, **193**, 113.
- Fayez, K. A. (1984): *Combined effect of sodium chloride and some nitrogen sources on growth and chemical composition of some plants*. M. Sc. Thesis, Assiut Univ. Sohag, Egypt.
- Heikal, M. M. (1975): Physiological studies on salinity. 1-Effect of saline irrigation on growth and photosynthetic pigments of safflower and sunflower plants. *Bull. Fac. Sci., Assiut University*, **4** (1), 1-11.
- Heikal, M. M. (1976): Physiological studies on salinity. 5-Effect of salinity on photosynthetic pigments and nitrogen content and on growth of wheat and radish plants. *Bull. Fac. Sci., Assiut University*, **5** (3), 243-256.
- Heikal, M. M., Ahmed, A. M., Zidan, A. M. (1979): Some physiological responses of *Phaseolus vulgaris* to different levels of sodium chloride. *Bull. Fac. Sci., Assiut University*, **8** (1), 159-175.
- Heikal, M. M., Ahmed, A. M., Shaddad, M. A. (1980a): Changes in dry weight and mineral composition of some oil producing plants over a range of salinity stress. *Biologia Plantarum (Praha)*, **22** (1), 25-33.
- Heikal, M. M., Ahmed, A. M., Shaddad, M. A. (1980b): Dry matter and mineral composition of some oil producing plants as influenced by some salinization treatments. *Phyton (Austr.)*, **20**, 159-173.
- Heikal, M. M., Ahmed, A. M., Shaddad, M. A. (1980c): Salt tolerance of some oil producing plants. *Agricultura (Heverlee)*, **22** (3), 437-453.
- Heikal, M. M., Ahmed, A. M., Zidan, M. A. (1981): Some physiological response of two cowpea cultivars to different levels of sodium chloride. *Bull. Fac. Sci., Assiut University* (in press).
- Helal, H. M., Mengel, K. (1979): Nitrogen metabolism of young barley plants as affected by NaCl salinity and potassium. *Plant and Soil*, **51**, 457-462.
- Ismail, A. M. (1982): *Studies on the effect of salinity on growth and chemical composition of some plants*. M. Sc. Thesis, Assiut University, Assiut, Egypt.
- Jacoby, B. (1965): Sodium retention in excised bean stem. *Physiol. Plantarum*, **18**, 730-739.
- Joshi, G. V., Naik, G. R. (1980): Response of sugarcane of different types of salt stress. *Plant and Soil*, **56**, 255-263.
- Lahaye, P., Epstein, E. (1969): Salt toleration by plants. Enhancement with calcium. *Science*, **166**, 395-396.
- Mehrotra, C. L. (1971): Salt tolerance of some agricultural crops during early growth stages. *Indian J. Agric. Sci.* **41**, 882.
- Metzner, H., Rau, H., Senger, H. (1965): Untersuchungen zur Synchronisierbarkeit einzelner-pigment-Mangel Mutanten von *Chlorella*. *Planta*, **65**, 186-194.
- Paech, K., Tracey, M. V. (1956): *Modern Methods of Plant Analysis*. Vol. 1. Springer-Verlag, Berlin.
- Patil, P. K., Patil, V. K., Ghonsikar, C. P. (1984): Effect of soil salinity on growth and nutritional status of guava (*Psidium guajava*). *Int. J. Trop. Agric.* **2**, 337-344.
- Radi, A. F. (1964): *Effect of environmental condition on the absorption and distribution of mineral elements in plants*. Ph. D. Diss. Moscow.
- Rush, D. W., Epstein, E. (1976): Genotypic responses to salinity. Differences between salt-sensitive and salt-tolerant genotypes of the tomato. *Plant Physiol.*, **157**, 162-166.
- Salama, F. M., Khodary, S. A., Heikal, M. M. (1980): Effect of saline irrigation and gibberellic acid on osmotic pressure, photosynthetic pigments and carbohydrates content of carrot and sugarbeet plants. *Egypt. J. Bot.*, **23**, 113-121.
- Schlegel, H. G. (1956): Die Verwertung organischer Säuren durch *Chlorella* in Licht. *Plant.*, **47**, 510.
- Schwarzenbach, G., Biederman, W. (1948): Komplexone X. Erdalkalikomplexe von 0.6-Dioxyazofarbstoffen. *Helv. Chem. Acta*, **31**, 678-687.
- Saddad, M. A., Heikal, M. M. (1982): Interactive effect of gibberellic acid and salinity on kidney bean. *Bull. Fac. Sci., Assiut University* (in press).
- Shimose, N. (1963): Physiology of salt injury in crops. 3. Salt tolerance of matrush plants. *J. Sci. Soil, Tokyo*, **34**, 147-149.
- Shimose, N. (1964): Physiology of salt injury in crops. Rice plants grown in K and Ca deficient solutions and in solutions with excessive amount of NaCl or sulphate. *J. Sci. Soil, Tokyo*, **35**, 148-151.

- Shimose, N. (1968): Salt tolerance of onion, celery, spinach, cucumber and kidney bean plants. *J. Sci. Soil. Tokyo*, **39**, 548-553.
- Shimose, N. (1973): Physiology of salt injury in crops. X. Effect of environmental conditions on the growth of rice plants grown in excess salt solutions. *Sci. Rept. Fac. Agr., Okayama University*, **41**, 69-78.
- Snedecor, I. W., Cochran, W. G. (1967): *Statistical methods*. The Iowa State University Press, Ames, Iowa.
- Todd, G. W., Basler, E. (1965): Fate of various protoplasmic constituents in droughted wheat plants. *Phyton*, **22** (1), 79-85.
- Varshney, K. A., Bajjal, B. D. (1979): Influence of hormonal treatment on chlorophyll retention in leaf discs from some salt stressed grasses. *Comp. Physiol. Ecol.*, **4** (2), 104-105.
- Wilson, J. R., Haydock, K. P., Robins, M. F. (1970): Response to salinity in glycine. 5. Changes in the chemical composition of three Australian species of *G. Wightii* (*G. Javanica*) over a range of salinity stresses. *Aust. J. Exp. Agric. Anim. Husb.*, **10**, 156-165.
- Yousif, H. Y., Bingham, F. T., Yermanos, D. M. (1972): Growth, mineral composition and seed oil of sesame (*Sesamum indicum* L.) as affected by NaCl. *Soil. Sci. Amer. Proc.*, **36**, 450-452.





## EFFECT OF THINNING AND HALVING ON GRAIN DEVELOPMENT IN *TRITICALE*

M. R. RAO and V. K. KHANNA

DEPARTMENT OF PLANT BREEDING, PANTNAGAR, INDIA

(Received 1 September 1987; accepted 7 March 1988)

The effect of thinning and halving on grain development was studied in two strains of triticale. If thinning was done early, i.e. on the 40th day after sowing, grain yield per year did not increase. Halving the ears decreased, the total weight per shoot. Mean dry weight per grain, number of grains, grains dry weight and chaff dry weight increased. The basal half of the intact ear and stem dry weight also increased.

**Keywords:** 'Triticale', anthesis, halving, photosynthesis, thinning

### Introduction

There is ample evidence that grain yield is controlled by the size and efficiency of the photosynthetic system (Thorne 1974), although treatments that decrease the supply of photosynthate to the grain by shading or defoliation do not always reduce yield proportionately because there is compensatory movement of carbohydrate from other organs to the grain (Wardlaw et al. 1965, Puckridge 1968, Bremner 1972). There is also evidence that the capacity of the grains to accumulate carbohydrate may be as important as photosynthesis in determining yield. Bingham (1969) showed that decreasing the number of grains in the ear increased the size of remaining grains, indicating a dependence of yield on the supply of photosynthate. But the increases in grain size only partly compensated for the missing grains, indicating that storage characteristics of the grain were also important.

In triticale, a man-made cereal crop which has been synthesized by crossing wheat (*Triticum* sp.) with rye (*Secale cereale*), there is a persistence of grain shrivelling that results in low grain test weights and low flour yields. Very little is known about the factors that control the size of the grain when the carbohydrate supply is ample. They probably act within the grain itself (Jenner and Rathjen 1972, Thorne 1974) and not in the phloem (Evans et al. 1970). The factors within the grain may involve the resistance to movement of sucrose between the vascular bundle and the endosperm cavity (Jenner 1974), or the cell division in the endosperm that occurs until about 14 day after anthesis (Wardlaw 1970, Evers 1970, Brocklehurst 1977). Another pos-



sibility is that growth substances, whose concentrations in the grain change during development, regulate the growth of the grain, possibly via effects on water content and volume of the grain (Radley 1976).

### Materials and methods

The two experiments reported here were planned to study the relative importance for final grain size in triticale of conditions which might alter the storage capacity of the grain and also the supply of carbohydrate during grain growth. In the first experiment the number of tillers was decreased before anthesis to improve early ear growth of the remaining shoots. This treatment also decreased the competition for light after anthesis. To distinguish between the pre- and post-anthesis effects, the carbohydrate supply per grain was increased still further by removing the top half of some ears soon after anthesis.

"UPT 79342" and "UPT 79343" were grown in plastic pots of 4.5 litres capacity containing 5 kg of soil to which an ample amount of fertilizer was added for thinning and halving experiments, respectively. First treatment was a combination of two shoot densities, i.e. thinned and unthinned; and the second treatment was a combination of two sizes of the ear, i.e. halved and intact ear. Pots were kept in the net house.

Seeds were sown at five-day intervals of 3 sowing dates with 4 replications, and the first sowing date was on 17th of December, 1986. Five seedlings per pot were maintained after sowing. New tillers were removed regularly. Ears were halved 5 days after anthesis.

#### *Thinning*

The time of anthesis was recorded daily. Samples were taken from each pot at intervals during growth. Only the main shoots and larger tillers which reached anthesis first were used. One main shoot ear was sampled from each pot at 18 days after anthesis and again at 23 days after anthesis in thinned and unthinned pots. The stems were left intact so that competition for light was changed minimally. The two basal grains in the four uppermost spikelets of the lower half of the ear, excluding the top spikelets, were selected for measuring fresh weight, dry weight and water content. At maturity, the remaining three main shoots from the thinned pots were harvested.

#### *Halving*

The time of anthesis was recorded daily and each ear was halved 5 days after its own anthesis date. Only the main shoot and the larger tillers, which reached anthesis first, were used.

### Results

There was no significant difference between the fresh weight, dry weight and water content of thinned and unthinned plants (Tables 1, 2 and 3). Thinning decreased total dry weight per shoot at maturity by 15.5% (Table 4), through a decrease in weight of all plant parts. Grain dry weight per ear decreased by 20% because there were five less grains per ear and mean dry weight per grain also decreased by 5% in contrast with the selected grains.

Halving the ears decreased the total weight per shoot because grain dry weight was only 71% of the grain weight of intact ears (Table 4). Mean dry weight per grain, number of grains, grain dry weight and chaff dry weight increased. The basal half of the intact ear and stem dry weight also increased by 4.6%.

**Table 1**

*Mean effects of thinning the plants on fresh weight of grains on different days after anthesis*

Plant density	Days after anthesis			Mean
	18	23	70	
Thinned	64.30	57.58	42.33	54.75
Unthinned	67.21	58.84	43.87	56.64
Mean	67.75	58.21	43.10	55.69
	Plant density	Days after anthesis	Plant density x Days after anthesis	
C. D. (5%)	NS	5.17	NS	
C. V. %	8.84			

**Table 2**

*Mean effects of thinning the plants on dry weight of grains on different days after anthesis*

Plant density	Days after anthesis			Mean
	18	23	70	
Thinned	27.16	32.41	37.50	32.36
Unthinned	28.24	32.70	38.49	33.15
Mean	27.70	32.56	38.00	32.75
	Plant density	Days after anthesis	Plant density x Days after anthesis	
C. D. (5%)	NS	3.07	NS	
C. V. (%) =	8.90			

**Table 3**

*Mean effects of thinning the plants on water content of grain on different days after anthesis*

Plant density	Days after anthesis			Mean
	18	23	70	
Thinned	37.13	25.16	4.95	22.42
Unthinned	38.95	26.13	5.37	23.48
Mean	38.04	25.65	5.16	22.95
	Plant density	Days after anthesis	Plant density x Days after anthesis	
C. D. (5)%	NS	3.58	NS	
C. V. (%) =	14.84			



**Table 4**  
*Percentage change by thinning and halving the plants on dry weight at maturity on the various plant parts (per shoot)*

Characters	Unthinned	Thinned	Percent change over unthinned	Intact	Halved	Percent change over intact
<i>Whole shoot</i>						
Total dry weight (g)	3.66±0.06	3.17±0.06	13.39	3.66±0.06	3.06±0.06	16.40
Stem dry weight (g)	1.57±0.04	1.52±0.04	3.19	1.53±0.04	1.60±0.04	4.58
Chaff dry weight (g)	0.69±0.03	0.49±0.03	28.99	0.62±0.02	0.40±0.02	35.49
Grain dry weight (g)	1.40±0.05	1.16±0.05	17.15	1.51±0.04	1.06±0.04	29.81
Number of grains	42.90±0.19	37.58±0.24	12.40	47.25±0.18	27.00±0.18	42.86
Dry weight per grain (mg)	32.00±0.14	30.00±0.17	6.25	31.00±0.16	39.00±0.17	25.81
<i>Lower half ear</i>						
Chaff dry weight (g)	0.41±0.02	0.36±0.02	12.20	0.38±0.02	0.40±0.02	5.27
Grain dry weight (g)	0.77±0.03	0.65±0.04	15.59	0.85±0.03	1.06±0.04	24.71
Number of grains	23.58±0.14	21.00±0.18	10.95	22.66±0.17	27.00±0.18	19.16
Dry weight per grain (ng)	32.00±0.16	31.00±0.16	3.13	37.00±0.14	39.00±0.17	5.41

### Discussion

Thinning relieved competition for light and presumably increased photosynthesis in the remaining shoots. Removal of late tillers as soon as they appeared ensured that the initial increase in light intensity was maintained. However, when plant thinning was carried out at anthesis to make more carbohydrate available for grain filling in the remaining ears, grain yield per ear did not increase. It can be argued, therefore, that grain yield probably was determined at least partly by a limited ear capacity. Plant thinning at earlier stages showed how the development of competition during the ear development period progressively reduced the potential capacity of the ear; the greater competition of higher plant populations accelerated this reduction in ear potential. Willey and Holliday (1971) reported that yield per ear in wheat decreased as thinning was delayed.

Anthesis starts about ten days later in triticale, compared to wheat, so our results regarding thinning differ from those of Martinez-Carrasco and Thorne (1979), who reported an increase in total dry weight of the wheat plants, when thinning was done 72 days after sowing. Our results show that if thinning is done early, i.e. on the 40th day after sowing, grain yield per ear does not increase. The results of Lupton and Pinthus (1969) in wheat showed that small tillers, which die without forming ears may contribute to the carbohydrate supply of the spike-producing shoots and thus to their potential grain yield. A positive relationship between the number of unproductive tillers per plant and increase in grain yield, resulting from the growth of more spikelets per spike, had previously been demonstrated in a study of winter and spring wheat varieties (Pinthus 1967). In our study the unproductive tillers had been removed much earlier.

Halving the ears decreased the total weight per shoot. Mean dry weight per grain, number of grains, grain dry weight and chaff dry weight increased; and the basal half of the intact ear and stem dry weight also increased. Removing the apical half of the ear would have increased the supply of assimilates to the remaining grains, unless the rate of photosynthesis was decreased, which is unlikely (Lupton 1966, Austin and Edrich 1975).

### References

- Austin, R. B., Edrich, J. (1975): Effect of ear removal on photosynthesis, carbohydrate accumulation and on the distribution of assimilated  $^{14}\text{C}$  in wheat. *Ann. of Bot.*, **39**, 141-152.
- Bennett, M. D. (1977): Heterochromatin, aberrant endosperm nuclei and grain shrivelling in wheat-rye genotypes. *Heredity*, **39**, 411-419.
- Bingham, J. (1969): The physiological determinants of grain yield in cereals. *Agric. Prog.*, **44**, 30-42.
- Bremner, P. M. (1972): Accumulation of dry matter and nitrogen by grains in different positions of the wheat ear as influenced by shading and defoliation. *Aust. J. Biol. Sci.*, **25**, 657-668.



- Brocklehurst, P. A. (1977): Factors controlling grain weight in wheat. *Nature*, **266**, 348-349.
- Evans, L. T., Dunstone, R. L., Rawson, H. M., Williams, R. F. (1970): The phloem of the wheat stem in relation to requirements for assimilate by the ear. *Aust. J. Biol. Sci.*, **23**, 743-752.
- Evers, A. D. (1970): Development of the endosperm of wheat. *Ann. of Bot.*, **34**, 547-555.
- Gustafson, J. P., Bennett, M. D. (1982): The effect of telomeric heterochromatin from *Secale cereale* on triticales. I. The influence of the loss of several blocks of telomeric heterochromatin on early endosperm development and kernel characteristics at maturity. *Canad. J. Genet. Cytol.*, **24** (1), 83-92.
- Jenner, C. F. (1974): An investigation of the association between the hydrolysis of sucrose and its absorption by grains of wheat. *Aust. J. Pl. Physiol.*, **1**, 319-329.
- Jenner, C. F., Rathjen, A. J. (1972): Limitation to the accumulation of starch in the developing wheat grain. *Ann. of Bot.*, **36**, 743-754.
- Lupton, F. G. H. (1966): Translocation of photosynthetic assimilates in wheat. *Ann. App. Biol.*, **57**, 355-364.
- Lupton, F. G. H., Pinthus, M. J. (1969): Carbohydrate translocation from small tillers to spike-producing shoots in wheat. *Nature*, **221**, 483-484.
- Martinez-Carrasco, R., Thorne, G. N. (1979): Physiological factors limiting grain size in wheat. *J. Exp. Bot.*, **30**, 669-679.
- Pinthus, M. J. (1967): Effect of 2-chloro-ethyl trimethyl ammonium chloride on the progenies of treated coheat plants. *J. Sci. Food. Agri.*, **18**, 386-387.
- Puckridge, D. W. (1968): Photosynthesis of wheat under field conditions. I. The interaction of photosynthetic organ. *Aust. J. Agri. Res.*, **19**, 711-719.
- Radley, M. (1976): The development of wheat grain in relation to endogenous growth substances. *J. Exp. Bot.*, **27**, 1009-1021.
- Seal, A. G., Bennett, M. D. (1985): *Effect of parental genotype on early seed development in wheat-rye hybrids*. In Genetics and breeding of triticales (Ed. by Bernard, M. and Bernard, S.), 161-168.
- Thorne, G. N. (1974): Physiology of grain yield of wheat and barley. *Rothamsted Exp. Stn. Rep.*, **2**, 5-25.
- Varghese, J. P., Lelley, T. (1983): Origin of nuclear aberrations and seed shrivelling in triticales. A re-evaluation of the role of C-heterochromatin. *TAG*, **66** (2), 159-167.
- Wardlaw, I. F. (1970): The early stages of grain development in wheat. Response to light and temperature in a single variety. *Aust. J. Biol. Sci.*, **23**, 765-774.
- Wardlaw, I. F., Cary, D. J., Anderson, M. J. (1965): The relative supply of carbohydrate and nitrogen to wheat grains, and as assessment of the shading and defoliation techniques used for these determinations. *Aust. J. Agri. Res.*, **16**, 893-901.
- Willey, R. W., Holliday, R. (1971): Plant population, shading, and thinning studies in wheat. *J. Agri. Sci., Cambridge*, **77**, 453-461.

## RESPONSES OF FIVE FORAGE CROPS TO TEMPERATURE AND SALT STRESSES AT GERMINATION

M. A. EL FAWAL and F. S. EL NAKHLAWY

AGRONOMY AND RANGE SCIENCE DEPARTMENT, COLLEGE OF AGRICULTURE  
AND VETERINARY MEDICINE, KING SAUD UNIVERSITY, QASSIM BRANCH,  
SAUDI ARABIA

(Received: 26 November 1987; accepted in revised form: 14, March 1988)

Germination and seedling characteristics of five forage crops, alfalfa (*Medicago sativa* L.), Egyptian clover (*Trifolium alexandrinum* L.), common vetch (*Vicia sativa* L.), barley (*Hordeum vulgare* L.), and Sudangrass (*Sorghum bicolor* (L.) Moench.) were studied as influenced by four concentrations of NaCl and Na<sub>2</sub>SO<sub>4</sub> as well as 3 levels of day/night temperature. For seed germination, barley showed the best adaptability for the different levels of salinity and temperature followed by alfalfa, common vetch, Egyptian clover and finally Sudangrass. 30/17 °C temperature gave the lowest percentage of seed germination except in case of Sudangrass. Alfalfa proved to be the highest tolerant crop in terms of seedling characteristics while common vetch was the lowest. The negative influence of NaCl on percentage of seed germination and seedling characteristics was significantly higher than Na<sub>2</sub>SO<sub>4</sub>. For both salts, the rate of seed germination significantly decreased at the highest concentration (15,000 ppm). Most of the interactions among the four variables are highly significant.

**Keywords:** forage crops, germination, salt stress

### Introduction

Salinity, temperature and drought stresses are the main limiting factors for crop production in arid and semiarid regions. Epstein et al. (1980) stated that the salinity of soils and irrigation water is a problem that restricts yield on 40,000 hectares of irrigated land, which is approximately one-third of the world's irrigated area.

Underground water is subjected to different levels of salinization, especially under arid conditions. Subsequently, the first surface layer of the soil frequently becomes more saline than the subsurface. Moreover, evaporation tends to reduce the moisture content in the surface layers, thereby aggravating the effects of salinity (Ayers, 1952). Conceivably, the seed will be placed in a more saline environment than the established plants.

Several workers reported that crop plants markedly differ in their salinity tolerance. Furthermore, germination and early seedling growth particularly are more sensitive than later stages of development (Kling, 1954; Bernstein and Hayward, 1958 and Sinha et al., 1982). Norlyn and Epstein (1984) indicated that tolerance of salinity at germination and emergence is a highly desirable trait. Therefore, the use of germination and emergence



as a first indicator of salt tolerance seems valid. For arid and semi-arid regions, Sinha et al. (1982) showed that quick-germinating species should have an advantage because of their ability to become rapidly established on saline soils, when temporarily relieved of excess salts by rains. Agricultural production in the Qassim area of Saudi Arabia is managed under arid conditions, and underground water is the main resource of irrigation.

The present investigation was conducted to elucidate the germination potentiality of 5 forage crops treated with 4 concentrations of sodium chloride and sodium sulphate at 3 levels of day/night temperatures. Percentages of each of seed germination and normal seedlings and the length of seedling were determined.

### Materials and methods

The present experiment was carried out using the seeds of 5 forage crops, namely, alfalfa (*Medicago sativa* L.), Egyptian clover (*Trifolium alexandrinum* L.), common vetch (*Vicia sativa* L.), barley (*Hordeum vulgare* L.) and Sudangrass (*Sorghum bicolor* (L.) Moench.). A split-split-split-plot design with 3 replications was used. The main treatments were 3 levels of day/night temperature i.e. 20/7, 25/12 and 30/17 °C. Two salts (NaCl and Na<sub>2</sub>SO<sub>4</sub>) were the sub-treatments. The sub-sub treatments were assigned for 4 salt concentrations (0; 5,000; 10,000 and 15,000 ppm). The above-mentioned 5 crops represented the sub-sub-sub treatments.

For each replicate, 30 seeds of each crop were placed on a filter paper in sterilized petri dishes. The treatments were daily moistened with their assigned salt concentration. The experiment was conducted in a controlled environment growth chamber. Percentages of seed germination and seedling characteristics were evaluated after 7 days of inhibition. Normal seedlings percentage and seedling length were expressed as a percent of their corresponding control (0.0 salt concentration) to account for differences in germination among species.

Since the germination results represent a binomial distribution, an arcsine square root transformation has been used to normalize the data before analysis (Steel and Torrie, 1981).

### Results and discussion

#### Seed Germination

Data presented in Table 1, 2, 3 and 4 revealed the different potentials of the studied forage crops with respect to seed germination under different environmental conditions.

Generally speaking, barley showed the best adaptability for the different levels of salinity and temperature, followed by alfalfa, common vetch, Egyptian clover and finally Sudan-grass.

For the influence of temperature on seed germination, it is worth mentioning that the studied forage crops differ in their heat requirements. Sudangrass is a warm season crop while the other 4 crops prefer a temperature season. The highest level of temperature in the present study (30/17 °C) gave the lowest percentage of seed germination, except in the case of Sudangrass. Stroganov (1962) indicated that in some cases high temperature had a promoting effect on germination of warmth-requiring plants under conditions of

Table 1

*Results of significance tests for the effect of temperature, salts and salt concentration on germination and seedling characteristics of 5 forage crops*

Source of Variation	DF	Germination %	No. of normal seedlings	Seedling length
<i>Main Plots</i>				
Blocks	2			
Temperatures (T)	2	**	NS	**
Error "a"	4			
<i>Sub-Plots</i>				
Salts (S)	1	**	**	**
TXS	2	NS	NS	NS
Error "b"	6			
<i>Sub-Sub-Plots</i>				
Concentrations (C)	3	**	**	**
TXC	6	**	**	**
SXC	3	**	**	**
TXSXC	6	MS	**	NS
Error "c"	36			
<i>Sub-Sub-Sub-Plots</i>				
Forage Crops (F)	4	**	**	**
TXF	8	**	**	**
SXF	4	**	NS	*
CXF	12	**	**	*
TXSXF	8	**	**	*
TXCXF	24	**	NS	NS
SXCXF	12	**	**	NS
TXSXCXF	24	**	NS	*
Error "d"	192			
Total	359			

\* Significant at the 0.05 level of probability.

\*\* Significant at the 0.01 level of probability.

NS Nonsignificant.

normal salinity, but this effect was reversed under high salinity. Dealing with alfalfa, Bula and Massengale (1972) found that a general increase in temperature, within limits, increased the rate of germination and emergence with significant differences among cultivars in rate of germination at different levels of temperature. The lowest level of temperature (20/7 °C) resulted in a significant increase in germination percentages for common vetch, Egyptian clover and alfalfa, while the highest germination of barley was obtained at the medium level (25/12 °C). El-Sharkawy and Springuel (1979) found that



Table 2

*Germination percentage of the 5 forage crops as influenced by 4 concentrations of NaCl and Na<sub>2</sub>SO<sub>4</sub> under 3 levels of day/night temperature*

Temperature °C	Salt	Salt concentration (ppm)	Germination (%)				
			Common Vetch	Barley	Egyptian clover	Alfalfa	Sudangrass
20/7	NaCl	0.0	100	83.3	70	90	70
		5.000	93.3	83.3	80	80	60
		10.000	93.3	73.3	73.3	76.7	30
		15.000	90	70	50	60	26.7
	Na <sub>2</sub> SO <sub>4</sub>	0.0	96.7	76.7	70	90	70
		5.000	96.7	86.7	70	93.3	73.3
		10.000	86.7	73.3	76.7	76.7	36.7
		15.000	86.7	86.7	66.7	93.3	43.3
25/12	NaCl	0.0	96.7	93.3	80	100	73.3
		5.000	80	93.3	80	83.3	70
		10.000	63.3	93.3	63.3	83.3	66.7
		15.000	10	86.7	0.0	6.7	56.7
	Na <sub>2</sub> SO <sub>4</sub>	0.0	100	98.3	75	90	76.7
		5.000	93.0	96.7	76.7	80	73.3
		10.000	70	90	76.7	76.7	66.7
		15.000	63.3	90	76.7	53.3	60
30/17	NaCl	0.0	86.7	80	80	80	96.7
		5.000	23.3	66.7	70	73.3	86.7
		10.000	20	60	63.3	73.3	66.7
		15.000	3.3	36.7	6.7	56.7	56.7
	Na <sub>2</sub> SO <sub>4</sub>	0.0	100	36.7	86.7	90	90
		5.000	86.7	80	76.7	83.3	83.3
		10.000	63.3	66.7	76.7	76.7	76.7
		15.000	20	56.7	66.7	76.7	63.3

the optimal germination temperatures were 20 and 28 °C for barley and Sudangrass respectively; while for Egyptian clover it was 20 °C (Dalianis, 1980).

The suppressive influence of NaCl on the percentages of seed germination was significantly higher than Na<sub>2</sub>SO<sub>4</sub> for all the studied crops at the different levels of temperatures. For both salts, the rate of seed germination significantly decreased at the highest concentration (15,000 ppm), except in the case of barley and alfalfa treated with Na<sub>2</sub>SO<sub>4</sub> at 20/7 °C and Egyptian clover at 25/12 °C. The studied crops varied obviously with respect to salinity tolerance at the different levels of temperature. For Sudangrass, the influence of high concentrations was more drastic at a low temperature (20/7 °C), yet this was true at a high temperature (30/17 °C) in the case of common vetch. Additionally, at the highest concentration (15,000 ppm), the studied crops gave different rates of seed germination.

Table 3

Effect of salt concentrations and temperature on number of normal seedlings and seedling length of the 5 forage crops expressed as a percent of their corresponding control (0.0 salt concentration)

Temperature °C	Salt	Salt concentration (ppm)	Normal seedlings					Seedling length				
			Common Vetch	Barley	Egyptian clover	Alfalfa	Sudangrass	Common Vetch	Barley	Egyptian clover	Alfalfa	Sudangrass
20/7	NaCl	0.0	100	100	100	100	100	100	100	100	100	100
		5.000	77.8	91	100	88.9	100	84.2	60	82.5	90.3	69.1
		10.000	63	50	94.4	66.7	13.3	63.9	29.6	48.1	52.2	0.0
		15.000	0.0	0.0	0.0	7.4	0.0	24.7	0.0	0.0	0.0	0.0
	Na <sub>2</sub> SO <sub>4</sub>	0.0	100	100	100	100	100	100	100	100	100	100
		5.000	100	100	100	100	83.3	99.5	73.2	95.3	100	103.7
		10.000	75.3	77.3	88.9	85.2	61.1	68.2	32.6	68.6	83.6	56.0
		15.000	10.7	68.2	44.4	81.5	16.7	55.3	12.7	34.8	61.0	0.0
	NaCl	0.0	100	100	100	100	100	100	100	100	100	100
		5.000	24.1	85.2	50	80	70	36.5	59.5	35.2	72.6	70.6
		10.000	10	29.7	41.6	80	36.7	23.9	0.0	27.1	58.2	41.7
		15.000	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
25/12	Na <sub>2</sub> SO <sub>4</sub>	0.0	100	100	100	100	100	100	100	100	100	100
		5.000	66.7	98.4	97.7	73.3	66.7	60.3	77.5	87.1	79.8	82.6
		10.000	23.3	84.7	97.7	66.7	56.7	25.5	48.9	60.3	63.3	60.3
		15.000	0.0	84.7	66.7	43.3	33.3	0.0	37.4	45.9	44.2	32.4
	NaCl	0.0	100	100	100	100	100	100	100	100	100	100
		5.000	0.0	83.3	68.1	87.5	88.5	24.4	79.9	85.9	89.4	88.0
		100.000	0.0	62.5	28	70	57.7	0.0	66.9	41.3	67.5	74.4
		15.000	0.0	0.0	0.0	12.5	19.2	0.0	0.0	0.0	17.7	37.1
	Na <sub>2</sub> SO <sub>4</sub>	0.0	100	100	100	100	100	100	100	100	100	100
		5.000	66.7	76	100	85.2	85.2	69.4	84.1	90.4	90.4	78.5
		10.000	33.3	60	87	70.3	63	38.5	63.8	73.1	73.1	65.8
		15.000	0.0	48	30.4	59.2	51.9	3.9	48.1	67.4	44.2	51.6
30/17	NaCl	0.0	100	100	100	100	100	100	100	100	100	100
		5.000	0.0	83.3	68.1	87.5	88.5	24.4	79.9	85.9	89.4	88.0
		100.000	0.0	62.5	28	70	57.7	0.0	66.9	41.3	67.5	74.4
		15.000	0.0	0.0	0.0	12.5	19.2	0.0	0.0	0.0	17.7	37.1
	Na <sub>2</sub> SO <sub>4</sub>	0.0	100	100	100	100	100	100	100	100	100	100
		5.000	66.7	76	100	85.2	85.2	69.4	84.1	90.4	90.4	78.5
		10.000	33.3	60	87	70.3	63	38.5	63.8	73.1	73.1	65.8
		15.000	0.0	48	30.4	59.2	51.9	3.9	48.1	67.4	44.2	51.6



**Table 4**  
*Values of L. S. D. at the 0.05 level of probability*

Treatments	Germination %	Normal seedlings	Seedling length
Temperature	4.57	16.70	5.63
Salt (S)	3.63	3.47	3.87
Concentration (C)	2.93	1.95	3.56
Forage Crops (F)	2.57	3.77	4.71
TXC	5.07	3.38	6.12
SXC	4.14	2.76	5.02
TXF	2.57	6.53	8.80
SXF	3.64	5.33	7.07
CXF	5.14	7.54	9.41
TXSXF	6.30	9.23	11.98
TXCXF	8.91	13.05	9.83
SXCXF	7.27	10.99	12.14
TXSXCXF	12.60	18.46	21.96

From the statistical point of view, all the interactions among the 4 variables are highly significant, except those between temperature and salts, and among temperatures, salts and concentrations (Table 1).

Working on alfalfa, Ismail (1983) showed that salinity affected the rate of germination adversely and the presence of NaCl in the germination culture inhibited germination significantly more or less than that due to moisture stresses. Also, Stone et al. (1979) stated that at lower osmotic potentials, the germination of alfalfa seeds declined sharply with highly significant interactions regarding temperature, osmotic potential and cultivars. On the other hand, low salt concentrations tend to stimulate germination of some alfalfa cultivars (Ahi and Powers, 1938, and Mulwani and Polland, 1939). Moreover, Croughan et al. (1978) hypothesized that salt-tolerant alfalfa lines performed poorly in the absence of NaCl and that a substantial amount of NaCl was required for optimal growth.

Dealing with sorghum, several workers concluded that it is a fairly tolerant crop under salinity, however, varieties differed in their ability to resist salt concentration (Kaliappan and Rajagopal, 1969; Padmanathan and Rao, 1975 and Ogra and Baijal, 1978). Referring to barley, Rathore et al. (1977) found that only 2 varieties, out of 22, had more than 50% germination at the salinity level of 40 mmol and the interaction between varieties and salinity was significant.



### *Seedling Characteristics*

As a matter of fact, studying only the percentage of seed germination is insufficient to evaluate the crop tolerance regarding the different stress conditions. Seedling characteristics are reliable traits in this respect. In all treatments, alfalfa proved to be the highest tolerant crop in terms of normal seedling percentage, while common vetch was the lowest (Tables 1, 2, 3 and 4). Increasing degrees of temperature led to a significant decreases of normal seedlings for common vetch, barley and Egyptian clover, while a significant increase was obtained for Sudangrass. In the case of alfalfa, however, the lowest percentage of normal seedlings was detected at the medium level of temperature (25/12 °C).

The suppressive effect of NaCl on the normal growth of seedlings was significantly higher than that of  $\text{Na}_2\text{SO}_4$  for all crops and at the different levels of temperature. In most cases, no normal seedlings were obtained at the highest concentration of NaCl. For  $\text{Na}_2\text{SO}_4$ , however, different percentages of normal seedlings were detected under the highest concentration in all crops, except common vetch. Tables (1) shows that the interactions are highly significant except TXS, SXF, TXCXF and TXSXCXF.

Seedling length was measured as a criterion for evaluation of seedling growth under different stresses. The present data clearly indicated that the growth of seedlings of all crops was generally affected by all the treatments; however, the studied crops behaved differently. Generally speaking, alfalfa ranked first in terms of seedling tolerance, followed by Sudangrass, Egyptian clover, barley and finally common vetch. With respect to the effect of temperature treatments, both alfalfa and common vetch showed the best tolerance at the lowest level (20/7 °C), while barley and Sudangrass gave the highest seedling lengths at the highest level (30/17 °C). For Egyptian clover, however, the lowest and highest levels of temperature gave significantly higher results for the seedling length.

It is worth mentioning that the growth of seedlings under NaCl treatment was significantly lower than under  $\text{Na}_2\text{SO}_4$  in most cases. Moreover, the growth of seedlings was obviously suppressed as the concentration of NaCl increased. For  $\text{Na}_2\text{SO}_4$ , however, the crops varied significantly. Alfalfa and Egyptian clover showed a relatively reasonable response to the highest concentration (15,000 ppm) of  $\text{Na}_2\text{SO}_4$  at the different levels of temperature. Barley and Sudangrass displayed the highest tolerance at the highest level of temperature (30/17 °C). Common vetch was obviously affected by the high concentrations at the medium and high levels of temperature. Working on sorghum, Padmanathan and Rao (1975), found that root length progressively lessened corresponding to increasing salinity levels. Seedling length was affected greatly corresponding to salt concentration. Ogra and Baijal (1978)



also concluded that coleoptile growth was more adversely affected and seemed to be a better index for salt tolerance at the early seedling stage. El-Sharkawy and Springuel (1979) indicated that radicle emergence decreased at osmotic water potential lower than  $-7$  and  $-5$  for barley and Sudangrass, respectively.

### Acknowledgements

The authors would like to acknowledge the assistance of Mr. M. Basyoni, lab. technician.

### References

- Ali, S. M., Powers, W. L. (1938): Salt tolerance of plants at various temperatures. *Plant Physiol.*, **13**, 767-789.
- Ayers, A. D. (1952): Seed germination as affected by soil moisture and salinity. *Agron. J.*, **44**, 82-84.
- Bernstein, L., Hayward, H. E. (1958): Physiology of salt tolerance. *A. Rev. Plant. Physiol.*, **9**, 25-46.
- Bula, R. J., Massengale, M. A. (1972): *Environmental Physiology*, 167-184. (In C. H. Hanson (ed.) *Alfalfa Science and Technology*.) Amer. Soc. Agron. Inc., Publisher.
- Croughan, T. P., Stavarek, S. J., Rains, D. W. (1978): Selection of a NaCl tolerant line of cultured alfalfa cells. *Crop Sci.*, **18**, 959-963.
- Dalianis, C. D. (1980): Effect of temperature and seed size on speed of germination, seedling elongation and emergence of berseem and Persian clovers. (*Trifolium alexandrinum* and *T. resupinatum*). *Seed Sci. et Technol.*, **8**, 323-331.
- El-Sharkawy, H. M., Springuel, I. V. (1979): Germination of some crop plant seeds under salinity stress. *Seed Sci. et Technol.*, **7**, 27-37.
- Epstein, E., Norlyn, J. D., Rush, D. W., Kingsbury, R. W., Kelley, D. B., Cunningham, G. A., Wrona, A. f. (1980): Saline culture of crops: a genetic approach. *Science (Washington DC)*, **210**, 399-404.
- Ismail, A. M. A. (1983): The effect of quality of irrigation water, salinity, moisture stress and kinetin on germination of three cultivars of alfalfa. *Arab Gulf J. Scient. Res.*, **1**, 569-581.
- Kaliappan, R., Rajagopal, A. (1969): Salinity effect on the germination and early vigour of five sorghum varieties. *Madras Agr. J.*, **56**, 282-285.
- Kling, E. G. (1954): Physiology of plants on saline soils. *Glarnyi Bot. Sad. Biul. (USSR)*, **1**, 59-73.
- Mulwani, B. T., Polland, A. G. (1939): Effects of alkali salts on germination of seeds. *Agric. Live-Stock of India*, **9**, 548-555.
- Norlyn, J. D., Epstein, E. (1984): Variability in salt tolerance of four triticale lines at germination and emergence. *Crop Sci.*, **24**, 1090-1092.
- Ogra, R. K., Baijal, B. D. (1978): Tolerance of some sorghum varieties to salt stress at early seedling stage. *Indian J. Agric. Sci.*, **48**, 713-717.
- Padmanathan, G., Rao, J. S. (1975): Effect of salinity on the germination and growth of sorghum varieties at seedling stage. *Madras Agric. J.*, **62**, 537-540.
- Rathore, A. K., Sharma, R. K., Lal, P. (1977): Relative salt tolerance of different varieties of barley (*Horeum vulgare* L.) at germination and seedling stage. *Annals of Arid Zone*, **16**, 53-60.
- Sinha, A., Gupta, A. R., Rana, R. S. (1982): Effects of osmotic tension and salt stress on germination of three grass species. *Plant and Soil.*, **69**, 13-19.
- Steel, R. G. D., Torrie, J. H. (1981): *Principles and procedures of statistics*. McGraw-Hill Book Company Inc., N. Y.
- Stone, J. E., Marx, D. B., Dohrenz, A. K. (1979): Interaction of sodium chloride and temperature on germination of two alfalfa cultivars. *Agron. J.*, **71**, 425-427.
- Stroganov, B. P. (1962): *Physiological basis of salt tolerance in plants*. Izdatei Sivo Akad. Nauk USSR.



## *Plant cultivation*

### SELECTION OF POLLINATING PLUM VARIETIES AND THEIR PLACEMENT IN THE ORCHARD

Z. SZABÓ and J. NYÉKI

UNIVERSITY OF HORTICULTURE AND FOOD INDUSTRY, BUDAPEST, HUNGARY

(Received 26 May 1987; accepted 10 September 1987)

The self-pollination of plum varieties is a variety trait which is slightly influenced by the growing site.

On the basis of phenological observations of flowering and of fertilization studies, the varieties "Cacanska leptica", "Cacanska rodna" and "Bluefre" proved to be satisfactory pollinators for the self-sterile variety *Cacanska najbolja*.

As pollinators for the self-sterile variety "President", "Bluefre" proved excellent and "Stanley" satisfactory.

**Keywords:** degree of clicking in degree of self-fertilization, fertilization of open pollinated flowers, flowering periods, flowering phenology, flowering order, flowering time groups, selection of pollinators for self-sterile and partially self-sterile varieties, variety associations

#### Introduction

The beginning of flowering is a variety trait, though the weather may produce greater differences in the date of beginning of flowering than are caused by the internal properties of the varieties. Considerable deviations can be observed yearly and from one variety to the other in the date when flowering begins, but the relative flowering order of plum varieties is almost identical each year (Tóth 1957).

According to Tóth (1957) cross pollination is possible if the flowering periods of the plum varieties coincide for at least three days even under unfavourable weather conditions.

The self-fertilization ability of plum varieties is a genetically determined, constant character. The advantages of varieties were summed up by Tóth (1969) as follows:

- good fertility,
- great yield stability,
- no need for pollinating varieties,
- pollen has good fertilizing ability,
- no mutual sterility within the group.



For varieties with greater flower density and larger fruit, even a lower rate of seed setting may result in a high yield. Despite relatively low fruit setting (between 7 and 13%) the variety "Zimmers Frühzwetsche" produces good yield (Lee and Bünnemann 1981).

In case of cross-pollination the lower limit for satisfactory fertilization is 10%. In order to ensure a stable yield for self-sterile plum varieties, at least two pollinating varieties should be chosen and these should be planted alternately. It is advisable to plant pollinating varieties in every 2nd-3rd rows (Tóth 1967).

Based on the ease with which plum varieties can be fertilized, following recommendations were made by Tóth (1980) on the ratio of varieties requiring pollen to pollen donor varieties female to pollinator varieties:

- when fertilization is easy: 4 : 1 : 1;
- when fertilization is moderately easy: 2 : 1 : 1;
- when fertilization is difficult: 1 : 1 : 1.

It is not advisable to plant blocks more than two rows deep in case of self-sterile plum varieties. In the third row from the pollinating variety (Tóth 1967) or at a distance of over 15 m (Keulemans 1980) a substantial yield reduction can be observed in self-sterile plum varieties. The greatest permissible distance from the pollinating variety is 16 m (Soltész 1979).

In examinations carried out by Nyéki et al. (1985) the self-sterile plum variety "President" was satisfactorily fertilized by the varieties "Bluefre" and Stanley. There was a high degree of clicking between the flowering periods of the pollinating varieties "Cacanska leptica", "Cacanska rodna", "Cacanski" hybrid 11/11/80/59 and "Stanley" and the self-sterile variety "Cacanska najbolja". Even in case of self-fertilizing plum varieties Tóth (1980) argues that yield stability can be increased by planting a mixture of 2-3 varieties.

The presence of bees in plum orchards is essential (Keulemans 1980), as this leads to an increase in yields even in orchards consisting of self-fertile plum varieties (Tóth 1969). Experience shows that 4-5 bee families are required per hectare to ensure satisfactory pollination (Tóth 1967).

### Material and methods

Experiments on flowering and fertilization of plum varieties were carried out on the Siófok State Farm, the Csány State Farm, the "Hungarian-Soviet Friendship" Cooperative Farm, Kecskemét, and the Szamosmenti State Model Farm, Fehérgyarmat. The trees were planted between 1978 and 1981 with a row and plant distance of  $7 \times 4$  m at Siófok and  $8 \times 5$  m at the other sites. All the trees were on myrobalan rootstocks except for the Siófok orchard, where both GF 31 and myrobalan rootstocks were used.

Observations of flowering dynamics were carried out on branch sections designated for studies on open pollination. The number of flowers which were open, or already over, was counted every one or two days. At the beginning of flowering 1-5% of the flowers were open, while at the end of flowering 95-100% of the flowers were over.



In order to determine the degree of open fertilization 100–500 flowers per variety were counted on branches in various parts of the crown.

For the study on self-fertilization branch sections were isolated in the white bud stage using parchment bags which were removed after once the flowers were over. In each variety, 100–300 flowers were examined. The pollen used in cross-pollinations was collected from branches forced to flower and the pollination was carried out at full flowering on 150–1000 flowers per variety on isolated branch sections. Fruit setting was evaluated three weeks after petal shedding, after the shedding of fruit in June, and prior to ripening. The fruit was considered to be ripe when it had reached the size, shape, colour and flavour characteristic of the variety.

## Results

Investigations on the introduction of plum varieties have been underway at the Fruit Growing Department since 1974. Part of this work involves a study of the flowering and fertilization properties of foreign varieties. It is important to select satisfactory pollinating varieties for self-sterile and partially self-fertile varieties.

The degree of self-fertilization of plum varieties is reviewed in Table 1. Observations carried out for 1–4 years showed the following varieties to be self-sterile or only partially self-fertile (less than 10% fruit setting): “Ruth Gerstetter”, “Early Laxton”, “Czar”, “Tuleu timpuriu”, “Zöld ringló”, “Cambridge Gage”, “Cacanska najbolja”, “Ageni”, “Debreceni muskotály”, “Bluefre”, “President”, “Reine-Claude de Bavay” (Table 2). With the exception of “Bluefre” these varieties require a pollinator in most years. As “Bluefre” has high flower density and large fruit it gives a plentiful yield even if fruit setting is only 7–10%. The varieties examined provide a good illustration of the fact that plum varieties may range from completely self-sterile (“Debreceni muskotály”) to varieties with excellent self-fertilization (“Besztercei Bt. 2.”).

The study of self-fertilization was carried out without pollination under isolator bags. Pollen could only reach the stigma due to air movements. Under natural conditions the degree of self-fertilization is greater due to pollination by insects.

The fertilization of free-standing flowers again shows great deviations from one variety to another. Low fruit setting of less than 10% was observed for the varieties “Ruth Gerstetter”, “Tuleu timpuriu” and “Cacanska najbolja” (Table 3).

The degree of self-fertilization and open fertilization also change substantially yearly. It can be observed that the open fertilization is also lower for self-sterile and partially self-fertile plum varieties.

After determining the fertilization ability of the varieties, pollinators must be chosen for the varieties with poor fertilization ability, taking into account the flowering periods. Groups of flowering periods were set up for the plum varieties (Table 4). Pollinating varieties for self-sterile varieties can be chosen out of those belonging to the same or neighbour group of flowering



Table 1

Self-fertilization of plum varieties (Siófok 1982/1985)

Variety	1982		1983		1984		1985		Mean fruit setting in the years examined (%)
	Isolated flowers (pc)	Ripe fruit (%)	Isolated flowers (pc)	Ripe fruit (%)	Isolated flowers (pc)	Ripe fruit (%)	Isolated flowers (pc)	Ripe fruit (%)	
Ruth Gerstetter					175	0	—	—	0
Early Laxton					123	0	210	1.9	1.0
Czar					112	4.5	178	5.6	5.1
Ontario	106	17.0	205	28.8	189	25.4	89	42.3	28.4
Tuleu timpuriu					375	0	259	0	0
Krikon					173	12.7	115	21.7	17.2
Early Italian	117	32.5	215	21.4	138	9.4	89	10.1	18.4
Richards Early Italian	123	22.0	203	19.8	169	6.5	153	5.2	13.4
Zöld ringló					203	3.0	228	0.9	2.0
Cambridge Gage					121	0	196	2.6	1.3
Cacanska najbolja					—	—	336	0	0
Olasz kék					173	17.3	134	19.4	17.3
Victoria					103	0.8	239	15.5	11.2
Korai besztercei					47	10.6	80	28.8	19.7
Besztercei Bt. 2.	143	11.2	144	13.2	122	36.1	221	42.5	25.8
Ageni					166	3.0	171	0.6	1.8
Pozegaca					112	22.3	101	23.8	23.1
Stanley			212	15.6	260	6.9	230	13.0	11.8
Besztercei Nm 122					96	31.3	99	43.4	37.4
Debreceni muskotály					82	0	108	0	0
Bluefre	160	3.8	213	7.5	194	7.7	447	10.7	7.4
President	242	0	208	0	151	0	264	0.4	0.1
Reine-Claude de Bavay					157	10.2	240	4.2	7.2
Grovers Late Victoria	121	16.5	230	6.0	—	—	153	8.5	10.3

Table 2

*Grouping of plum varieties according to the extent of self-fertilization  
(Siófok, 1982–1985)*

Completely self-sterile (fruit setting 0%)	Partially self fertile (fruit setting between 0.1–10%)	Self-fertile (fruit setting over 10%)
Ruth Gerstetter	Early Laxton	Ontario
Tuleu timpuriu	Czar	Krikon
Cacanska najbolja	Zöld ringló	Early Italian
Debreceni muskotály	Cambridge Gage	Richards Early Italian
President	Ageni	Olasz kék
	Bluefre	Victoria
	Reine-Claude de Bavay	Korai besztercei
		Besztercei Bt. 2.
		Pozegaca
		Stanley
		Besztercei Nm 122.
		Grovers Late Victoria

periods groups. Compared to the early flowering varieties, flowering in the intermediate group began 2–3 days later in 1984 and 1–2 days later in 1985 and that the late flowering varieties 4–5 days late in 1984 and 3–4 days later in 1985. The date and order of flowering for the plum varieties are illustrated in Figs 1 and 2. There is very slight difference in the order of flowering for the two years in question. The flowering period of “Tuleu timpuriu” was somewhat unstable, as it flowered much earlier in 1984 than in 1985, compared to the other varieties.

Among the widespread or promising plum varieties, the flowering period of “Bluefre” was nearest to that of the self-sterile variety “President”. The coincidence of the flowering periods is well illustrated by the flowering phenograms (Fig. 3). In the four years examined the flowering of “Bluefre” coincided to the greatest extent with that of “President”. It can be observed that the course of flowering is influenced by the carying weather conditions in various years. In 1984 the low temperature experienced during the flowering period prolonged flowering, leading to distinct differences in the flowering periods of the individual varieties. In 1985 the weather was warmer at the beginning of flowering, so there were no great differences in the dates when flowering began, and the flowering periods coincided to a considerable extent. The early flowering variety “President” finished its flowering very quickly. The flowering of “Bluefre”, “Stanley” and “Besztercei Bt. 2” was prolonged by a drop in the temperature.

The extent of clicking in the flowering times of the plum varieties is also presented in tabular form (Tables 5 and 6). The high degree of coinci-



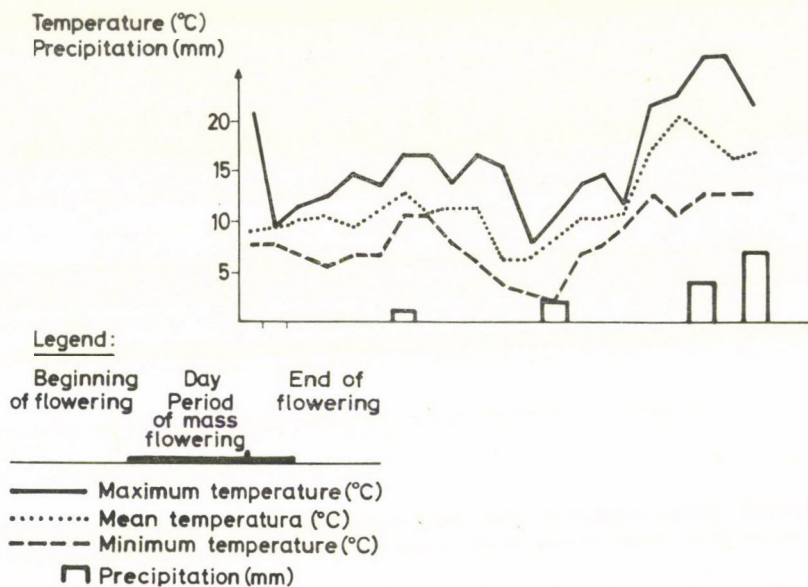
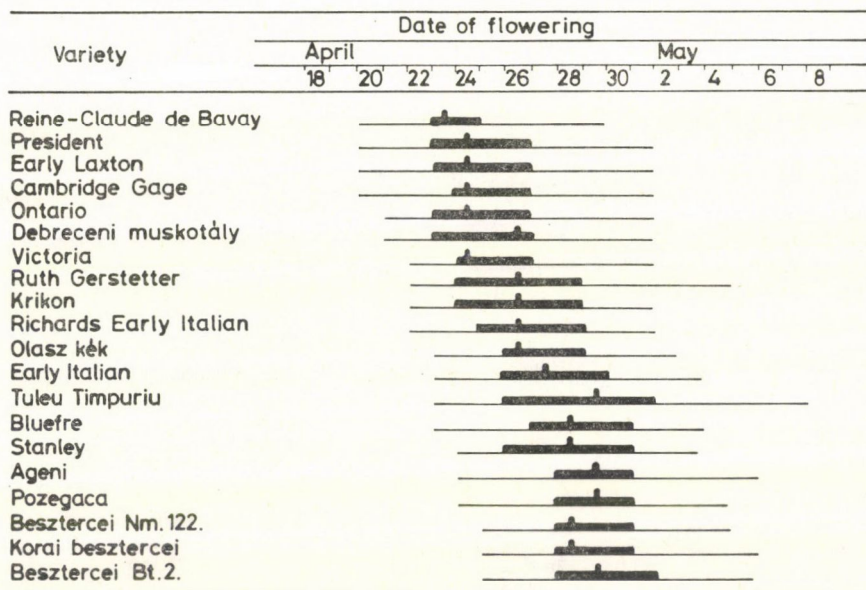


Fig. 1. Date and sequence of flowering for plum varieties (Siófok, 1984)

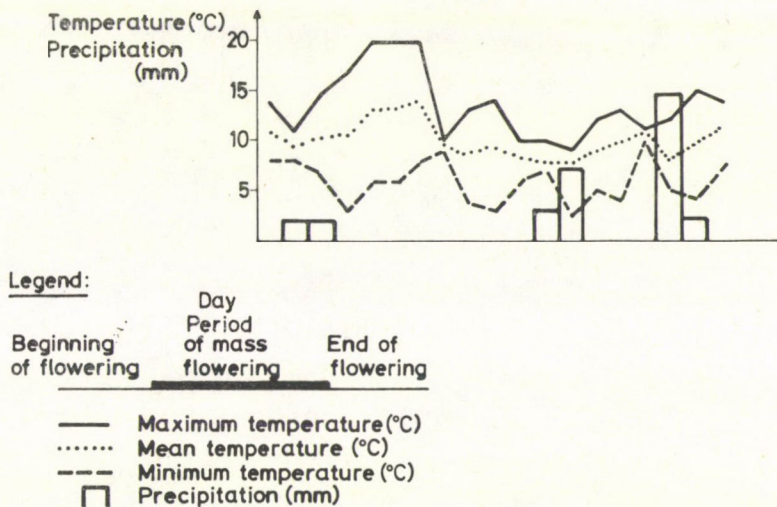
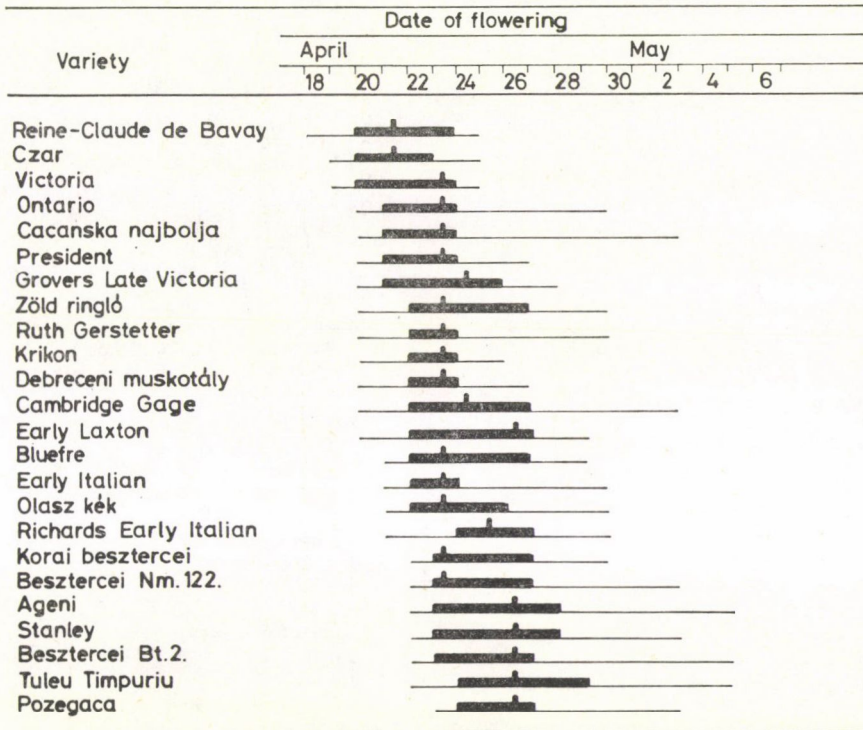


Fig. 2. Date and sequence of flowering for plum varieties (Siófok, 1985)



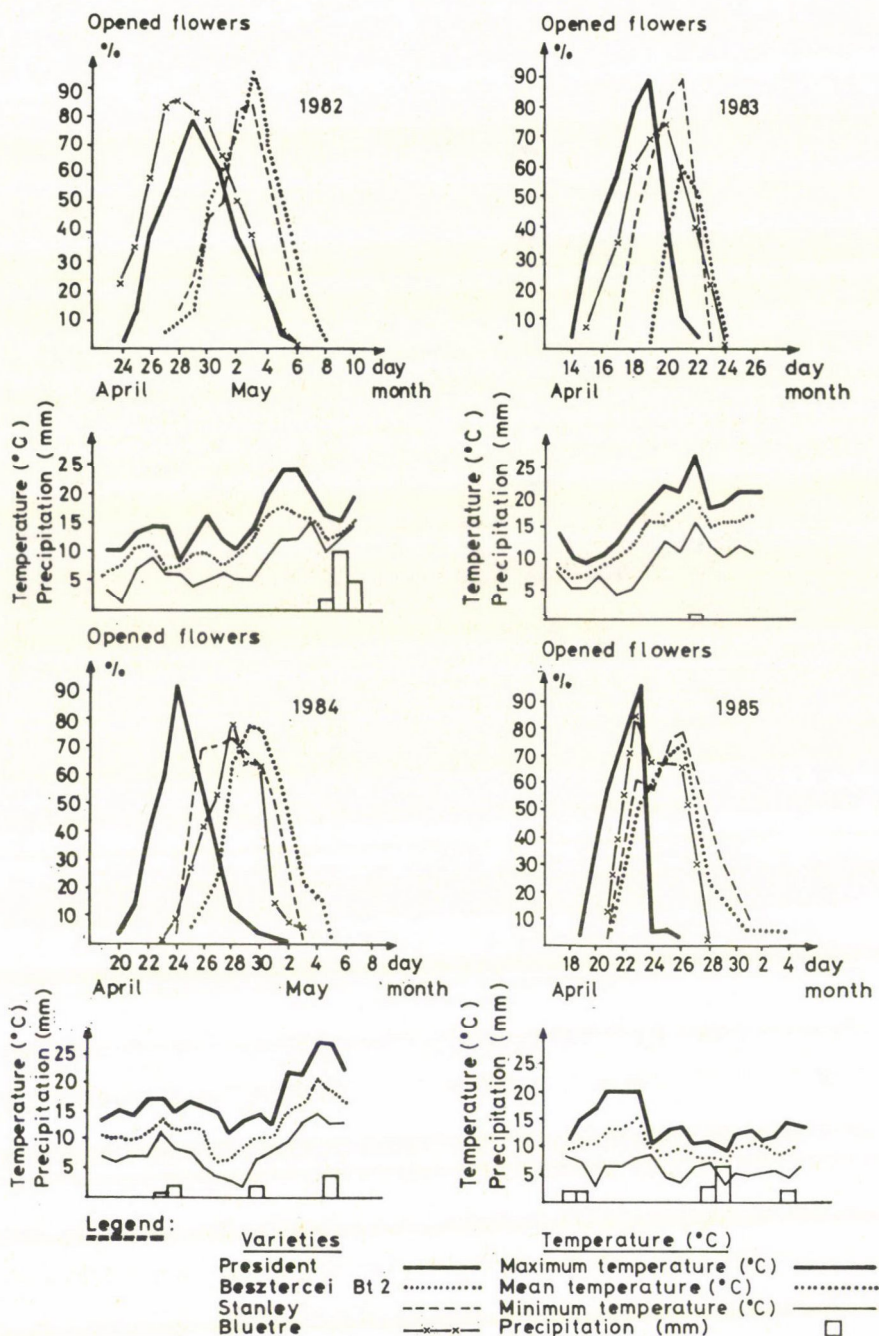


Fig. 3. Flowering phenograms of plum varieties (Siófok)

Table 3

*Open fertilization of plum varieties (Siófok. 1982-1985)*

Variety	1982		1983		1984		1985		Mean fruit setting in the years examined (%)
	Flowers observed (pc)	Ripe fruit (%)	Flowers observed (pc)	Ripe fruit (%)	Flowers observed (pc)	Ripe fruit (%)	Flowers observed (pc)	Ripe fruit (%)	
Ruth Gerstetter					141	9.9	1621	2.2	6.1
Early Laxton					268	6.3	1087	19.8	13.1
Czar					238	14.7	606	15.0	14.9
Ontario	171	52.0	274	48.9	178	39.9	311	24.1	41.2
Tuleu timpuriu					317	1.6	571	0.2	0.9
Krikon					73	20.5	381	27.3	23.9
Early Italian	158	45.6	220	32.0	206	23.8	248	2.8	26.1
Richards Early Italian	229	22.3	309	26.5	213	27.7	341	1.2	19.4
Zöld ringlő					289	12.1	240	12.1	12.1
Cambridge Gage					143	29.4	609	10.5	20.0
Cacanska najbolja					—	—	6043	5.0	5.0
Olasz kék					177	24.3	423	6.4	27.3
Victoria					214	42.5	484	27.7	35.1
Korai besztercei					139	10.1	139	30.2	20.2
Besztercei Bt. 2.	283	41.0	208	32.5	128	65.6	340	49.7	47.2
Ageni					80	28.8	786	16.4	22.6
Pozegaca					87	28.7	309	29.1	28.9
Stanley	203	30.0	305	23.6	269	37.5	643	14.5	26.4
Besztercei Nm 122					219	58.9	203	51.7	55.3
Debreceni muskotály					154	11.7	213	20.2	16.0
Bluefre	269	12.3	522	30.3	527	19.9	1656	15.5	19.5
President	335	19.4	319	25.7	390	7.4	1002	15.9	17.1
Reine-Claude de Bavay					176	13.6	585	14.4	14.0
Grovers Late Victoria	190	35.3	303	41.3	—	—	314	16.6	31.3



**Table 4**  
*Flowering time groups for plum varieties (Siófok, 1982–1985)*

Early flowering	Medium flowering	Late flowering
Early Laxton	Ruth Gerstetter	Tuleu timpuriu
Czar	Krikon	Korai besztercei
Ontario	Early Italian	Besztercei Bt. 2.
President	Richards Early Italian	Agén
Reine-Claude de Bavay	Zöld ringló	Pozegaca
Grovers Late Victoria	Cambridge Gage	Stanley
	Cacanska najbolja	Besztercei Nm. 122
	Olasz kék	
	Victoria	
	Debreceni muskotály	
	Bluefre	

**Table 5**  
*Clicking in the flowering times of the self-sterile plum variety "Cacanska najbolja" (♀) and that of possible pollinators*

Pollinating variety (♂)	Extent of clicking in flowering times									
	Fehérgyarmat		Siófok		Csány				Kecskemét	
	1983	1984	1985	1986	1983	1984	1985	1986	1985	1986
Ruth Gerstetter	—	—	77	—	—	—	—	—	—	—
Cacanska lepotica	100	100	—	—	83	93	92	100	100	75
Cacanska rodna	100	92	—	—	100	93	75	71	80	75
Stanley	100	77	85	75	100	100	75	86	90	—
Bluefre	—	—	62	100	—	—	—	—	80	88
Besztercei szilva	60	62	85	75	—	79	67	71	—	—
President	—	—	54	100	—	—	—	—	60	50

**Table 6**  
*Clicking in the flowering times of the self-sterile plum variety "President" (♀) and that of possible pollinators*

Pollinating variety (♂)	Extent of clicking in flowering times (%)						
	Siófok					Kecskemét	
	1982	1983	1984	1985	1986	1985	1986
Stanley	64	50	64	71	60	63	—
Bluefre	100	80	73	86	80	100	71
Besztercei szilva	67	40	55	71	60	—	—
Cacanska najbolja	—	—	—	100	80	75	57
Cacanska lepotica	—	—	—	—	—	75	86

**Table 7**  
*Cross-pollination of the self-sterile plum variety "Cacanska najbolja" ( $\delta$ )*

Pollinating variety (σ')	Siófok		Csány				Kecskemét		Mean fruit setting (%)
	1985		1985		1986		1986		
	Pollinated flowers (pc)	Ripe fruit (%)	Pollinated flowers (pc)	Ripe fruit (%)	Pollinated flowers (pc)	Ripe fruit (%)	Pollinated flowers (pc)	Ripe fruit (%)	
Ruth Gerstetter	95	7.4	140	7.8	—	—	—	—	7.6
Cacanska lepotica	83	45.8	88	20.4	818	6.0	145	2.1	18.6
Cacanska rodna	79	49.4	290	24.1	644	12.0	172	8.7	23.6
Stanley	206	3.9	319	9.1	1059	8.7	89	0	5.4
Bluefre	140	47.1	308	24.7	1239	11.2	131	6.9	22.5
Besztercei szilva	44	2.3	385	31.4	513	8.6	114	7.0	12.3
President	197	34.5	445	33.0	974	3.4	118	2.5	18.4
Open fertilization of Cacanska najbolja	6043	5.0	16654	1.1	12197	1.4	1416	4.9	3.1

**Table 8**  
*Cross pollination of the self-sterile plum variety "President" ( $\beta$ )*

Pollinating variety (♂)	Siófok						Kecskemét				Mean fruit setting(%)
	1983		1984		1985		1985		1986		
	Pollinated flowers (pc)	Ripe fruit (%)	Pollinated flowers (pc)	Ripe fruit (%)	Pollinated flowers (pc)	Ripe fruit (%)	Pollinated flowers (pc)	Ripe fruit (%)	Pollinated flowers (pc)	Ripe fruit (%)	
Stanley	119	28.6	166	16.3	139	21.6	236	9.7	150	1.3	15.5
Bluefre	146	42.5	121	15.7	153	32.7	277	26.4	80	32.5	30.0
Besztercei szilva	174	8.0	164	20.7	15	6.7	119	33.6	93	15.1	16.8
Cacanska najbolja					68	19.1	196	5.1			12.1
Cacanska lepotica									41	14.6	14.6
Open fertilization of President	319	25.7	390	7.4	1002	15.9	724	28.6	280	41.1	23.7



**Table 9**  
*Fertilization ability of pollinating plum varieties (Siófok, 1983–1985)*

Pollinating variety (♂)	1983				1984				1985				Mean fertilizing ability (%)
	Pollinated variety (♀)	Pollinated flowers (pc)	Fruit setting (%)	Pollinated variety (♀)	Pollinated flowers (pc)	Fruit setting (%)	Pollinated variety (♀)	Pollinated flowers (pc)	Pollinated variety (♀)	Pollinated flowers (pc)	Fruit setting (%)	Fruit setting (%)	
President				Bluefre	94	24.5	Cacanska najbolja	197	Cacanska najbolja	197	34.5	34.5	
				Stanley	78	32.1	Bluefre	54	Bluefre	54	35.2	35.2	
				Besztercei Bt. 2.	52	42.3	Besztercei Bt. 2.	20	Stanley	20	35.0	35.0	
Mean						32.9		17	Besztercei Bt. 2.	17	52.9	52.9	
											39.4	39.4	36.2
Bluefre	President	146	42.5	President	121	15.7	Cacanska najbolja	140	Cacanska najbolja	140	47.1	47.1	
				Stanley	81	30.9	President	153	President	153	32.7	32.7	
				Besztercei Bt. 2.	62	30.6	Stanley	12	Stanley	12	0	0	
Mean			42.5			25.7	Besztercei Bt. 2.	12	Besztercei Bt. 2.	12	20.0	20.0	
											32.5	32.5	33.6
Stanley	President	119	28.6	President	166	16.3	Cacanska najbolja	206	Cacanska najbolja	206	3.9	3.9	
				Bluefre	94	21.3	President	139	President	139	21.6	21.6	
				Besztercei Bt. 2.	75	49.3	Bluefre	81	Bluefre	81	13.6	13.6	
Mean			28.6			28.9	Besztercei Bt. 2.	15	Besztercei Bt. 2.	15	26.7	26.7	
											16.5	16.5	24.7
Besztercei Bt. 2.	President	174	8.0	President	164	20.7	Cacanska najbolja	44	Cacanska najbolja	44	2.3	2.3	
				Bluefre	96	16.7	President	15	President	15	6.7	6.7	
				Stanley	74	20.3	Bluefre	17	Bluefre	17	5.9	5.9	
Mean			8.0			19.2	Stanley	11	Stanley	11	0	0	
											3.7	3.7	10.3

Table 10

*Fertilization ability of pollinated plum varieties (Siófok, 1983–1985)*

Pollinated variety (♀)	1983			1984			1985			Mean fertilizing ability (%)
	Pollinating variety (♂)	Pollinated flowers (pc)	Fruit setting (%)	Pollinating variety (♂)	Pollinated flowers (pc)	Fruit setting (%)	Pollinating variety (♂)	Pollinated flowers (pc)	Fruit setting (%)	
President	Bluefre	146	42.5	Bluefre	121	15.7	Bluefre	153	32.7	
	Stanley	119	28.6	Stanley	166	16.3	Stanley	139	21.6	
	Besztercei Bt. 2.	174	8.0	Besztercei Bt. 2.	164	20.7	Besztercei Bt. 2.	15	6.7	
							Cacanska najbolja	68	19.1	
Mean			26.4			17.6			20.0	21.3
Bluefre				President	94	24.5	President	54	35.2	
				Stanley	94	21.3	Stanley	81	13.6	
				Besztercei Bt. 2.	96	16.7	Besztercei Bt. 2.	17	5.9	
Mean						20.8			18.2	19.5
Stanley				President	78	32.1	President	20	35.0	
				Bluefre	81	30.9	Bluefre	12	0	
				Besztercei Bt. 2.	74	20.3	Besztercei Bt. 2.	11	0	
Mean						27.8			11.7	19.9
Besztercei Bt. 2.				President	52	42.3	President	17	52.9	
				Bluefre	62	30.6	Bluefre	12	50.0	
				Stanley	75	49.3	Stanley	15	26.7	
Mean						40.7			43.2	42.0
Cacanska najbolja							President	197	34.5	
							Bluefre	140	47.1	
							Stanley	206	3.9	
							Besztercei Bt. 2.	44	2.3	
Mean									22.0	



dence in the flowering periods is also obvious from the data in the tables. This coincidence changes from year to year; if a variety is to be a good pollinator there must be around 70% clicking in the flowering periods every year. During the years examined the flowering of the varieties "Ruth Gerstetter", "Cacanska lepotica", "Cacanska rodna", "Stanley" and "Bluefre" coincided well with that of "Cacanska najbolja", while in an old orchard "Stanley"

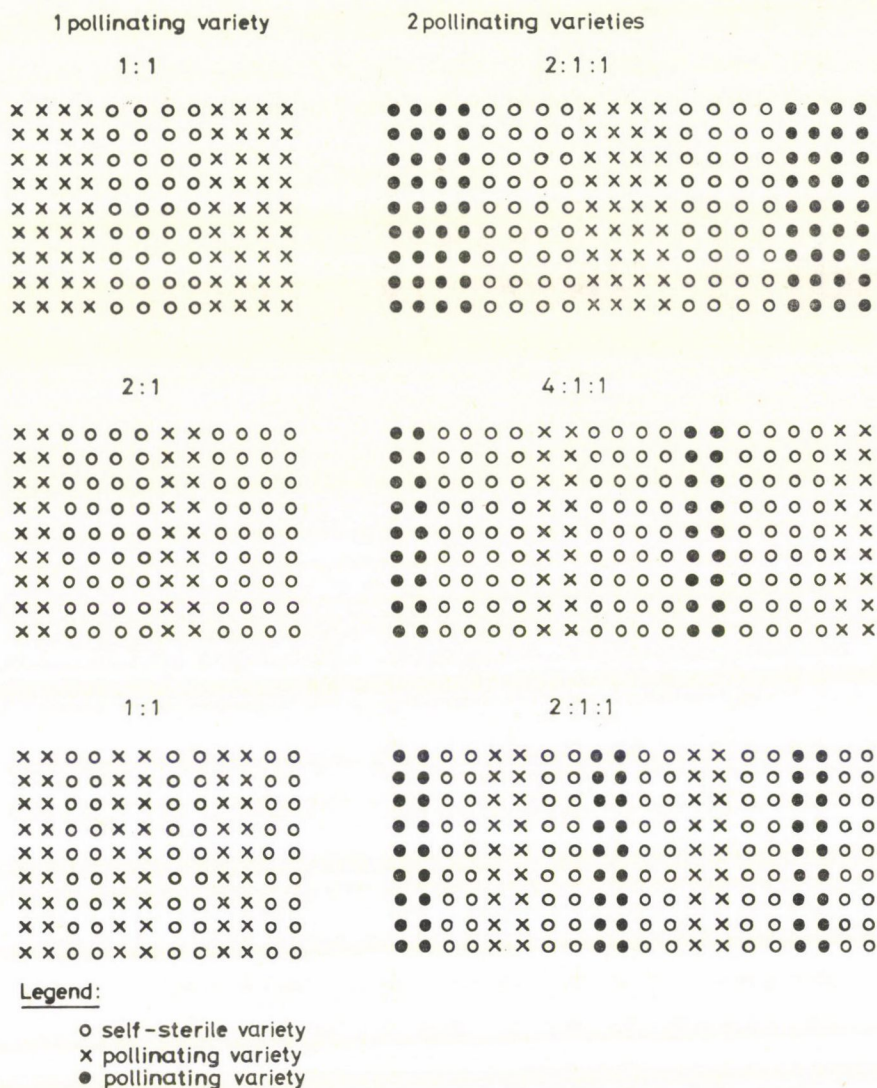


Fig. 4. Map of planting of plum variants

**Table 11**  
*Ripening time and utilization of plum varieties*

Variety	Ripening time	Utilization
Cacanska lepotica	Late July-early Aug.	Fresh consumption
Cacanska najbolja	Mid-August	Fresh consumption
Cacanska rodna	Second half of Aug.	Bottling, fresh consumption
Stanley	Late Aug.-early Sept.	All purpose
Bluefre	Late Aug.-early Sept.	Fresh, prunes
President	Early-middle Sept.	Fresh, prunes

flowered 1–2 days later. Thus, when determining the flowering period, the age of the orchard must also be taken into consideration.

Clicking with the flowering time of the variety "President" is greatest for "Bluefre" and "Cacanska lepotica" and less for "Stanley", "Besztercei" plum and "Cacanska najbolja".

Before choosing pollinating varieties their fertilizing ability must be determined first. Tables 7 and 8 contain the results of cross-pollinations. "Cacanska najbolja" was well fertilized in all the experimental years by "Cacanska rodna" and "Bluefre"; while "Ruth Gerstetter", "Cacanska lepotica", "Stanley", "Besztercei" plum and "President" were less satisfactory, though even for these varieties the mean fruit setting exceeded the degree of open fertilization.

The best pollinator for "President" was "Bluefre". The mean fruit setting exceeded 10% for all the varieties, but pollinations carried out with "Stanley", "Besztercei szilva" and "Cacanska najbolja" did not reach this figure in all years or at all growing sites.

The fertilizing ability of the pollen from plum varieties used as pollinators is shown in Table 9, and the ability of the flowers to become fertilized in Table 10. The pollen of "President", "Bluefre" and "Stanley" was highly fertile, while that of "Besztercei Bt. 2" gave relatively poor fertilization in 1986. The flowers of "Besztercei Bt. 2" were exceptionally fertile, while 20% of the flowers of "President", "Bluefre", "Stanley" and "Cacanska najbolja" were fertilized.

The variety associations within the orchard are determined by the fertilization abilities of the varieties. The placement problem of self-sterile and partially self-fertile varieties within the orchard can be solved by using one of the variants presented in Fig. 4. From the point of view of pollination it is better if only two rows of the self-sterile varieties are adjacent and several pollinators are used. The plantation of single rows is not recommended for labour organizational reasons.



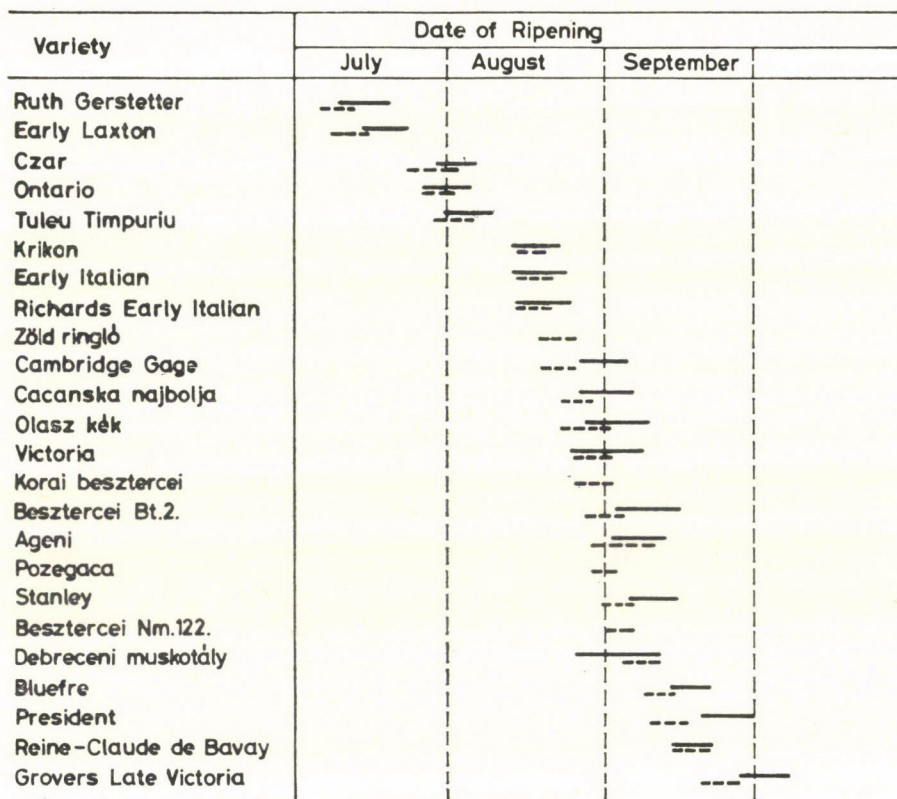


Fig. 5. Ripening of plum varieties (Siófok, 1984–1985)

The association of self-fertile plum varieties may also lead to increase in yield.

Economic considerations are the decisive factors in species and variety associations within the farm. The cultivation of fruit species and varieties which ripen one after the other prolongs the harvesting period, resulting in a less intense peak in labour requirements and a better utilization of machinery.

The ripening time of the plum varieties examined at Siófok are shown in Fig. 5. These varieties ripen over a period of three months, from mid-July to the middle or end of September. The plum varieties authorized for propagation and having high production and marketing value ripen in the order given in Table 11. If the ripening period is also taken into consideration, the self-sterile variety "Cacanska najbolja" can be recommended for association with "Cacanska leptica" and "Cacanska rodna", and the self-sterile variety "President" for association with "Stanley" and "Bluefre", using the plantations systems illustrated in Fig. 4.

### References

- Nyéki, J., Szabó, Z., M. Bóné-Tóth F., Pete, A. (1984): Flowering and fertilization of plum varieties. *Kertgazdaság*, 17 (2), 35–53.
- Soltész, M. (1979): *Variety associations in orchards*. (In: Gyuró, F., P. Vig (Eds): *Guidelines for orchard planning. Putting today's novelties into practice.*) Agricultural Press, Budapest, 52–55.
- Tóth, E. (1957): Comparative physiological and morphological studies on plum varieties. *Kertészeti Kutató Intézet Évkönyve*, 1–57.
- Tóth, E. (1967): The best pollinators for self-sterile plum varieties. Major results in agricultural research in 1966, 124–129.
- Tóth, E. (1969): *Study of the self-fertilization of plum varieties*. Thesis. (Manuscript) University of Horticulture, Budapest.
- Tóth, E. (1980): *Plums*. (In: Nyéki J. (Ed.): *Flowering biology and fertilization of fruit varieties.*) Agricultural Press, Budapest, 234–247.





## EFFECT OF DOLOMITE- AND LIME TREATMENT ON SOME QUALITY PARAMETERS OF JONATHAN APPLES

J. PAPP and A. H. AZIZ

UNIVERSITY OF HORTICULTURE AND FOOD INDUSTRY, DEPARTMENT OF POMOLOGY,  
BUDAPEST, HUNGARY

(Received 30 July 1988; accepted 10 August 1988)

On a sandy soil of poor magnesium status the effect of dolomite- and limestone dust treatments on the skin colour, firmness of flesh, total sugar- and acid content and Thiault's quality index of apples was studied at the time of harvest and at the end of storage.

Treatments with 2 t/ha and 4 t/ha dolomite and with limestone dust deepened the skin colour of the apples. In most years of the experiment the dolomite- and limestone dust treatments also increased the firmness of the apples compared to the control. No tendency-like effect of treatment on the total sugar content of the apples could be observed. In the majority of the experimental years the 4 t/ha quantity of dolomite and the lime treatment increased the acid content of the apples by the time of harvest, but at the end of storage the acid content both in the dolomite- and liming treatment decreased at a higher rate than in the untreated control.

On the average of the experimental years the apples in the 4 t/ha dolomite- and in the liming treatments showed significantly higher quality index values at the time of harvest.

There was no close correlation between the Mg content of the soil and the flesh firmness, acid content and quality index of the fruits. A significant correlation was found, on the other hand, between the Mg content of the soil and the Mg- and Ca contents of the apples.

**Keywords:** apple, dolomite- and lime fertilization, fruit quality data, Mg-supply

### Introduction

Deficient magnesium supply in apple orchards occurs more and more often (Trocmé 1960, Allen 1980). The deficient magnesium supply of fruit-trees must be reckoned with first of all on acidic sandy soils (Boynton-Oberly 1966, Van der Boon et al. 1966, Sadowski et al. 1976b, Papp 1988).

Although the natural magnesium status of the soil is ever more frequently found to have deteriorated due to the increased intensity of fruit production, the "relative Mg deficiency" caused by an overapplication of potassium is at least as important as that (Bergmann 1983, Sadowski et al. 1976a).

The use of dolomite (a limestone containing magnesium) for the improvement of the Mg supply of fruits has been studied for some time. A review of the subject was published by Boynton-Oberly (1966). The chemical activity of the different dolomite grists varies with their geological origin. According to Whitear (1978) the dolomite is suitable to form Mg reserves in acidic soils,



because it dissolves slowly and has a long-term effect. Lord (1983) suggests using dolomite to maintain the level of Mg in soils of orchards.

Highly acidic sandy soils are usually also highly Mg deficient; therefore when liming them a meliorative quantity of Mg is necessary, and dolomite is suitable for this purpose (Balogh et al. 1984). Sadowski et al. (1976b) found that the dolomite when applied in a quantity of 2.25 t/ha dissolved at a relatively fast rate on acidic sandy soil. While according to Kim-Lee (1980) 2.5 t/ha and 5.0 t/ha quantities of dolomite significantly increased the Mg content in apple leaves, Fisher et al. (1958) and Dione et al. (1967) found that the use of dolomite only slightly raised the Mg contents of leaves.

In most experiments carried out so far Mg fertilization did not significantly increase the yield (Hansen 1975, Sadowski 1976b, Pedersen-Vang Petersen 1984). Mg fertilization did not reliably influence the firmness of apple nor the skin colour (Sadowski et al. 1976b, Kim-Lee 1980). According to Ford (1968) in response to spraying with  $\text{MgSO}_4$  the number of bitter pit apples increased. Conversely in the experiments of Kim-Lee (1980), 5 t/ha applications of dolomite decreased the proportion of bitter pit fruits. Bergmann (1983) pointed out that in the case of Mg deficiency the Cox orange apple contained less sugar and showed poor storability.

### Materials and methods

The conditions of the experiment were described in our previous publication (Papp 1988).

On the basis of the visual examination of the effect of treatments on the skin colour of the apples we set up two categories (colour above and below 50%). The firmness of apple was determined using a manual fruit resistance tester on two occasions: on harvesting, and on the removal of the apples from the store-room. The data on the total sugar- and acid contents of the fruits as well as on the "quality index" values calculated by the method of Thiault (1970) were also obtained on these two occasions. The fruits were kept in a store-room with unchanged air space.

Bifactorial correlation examinations were carried out to clarify the relationship between the available Mg content in the 0-60 cm soil layer and the following factors:

- Mg content of leaves
- Ca content of leaves
- Mg content of fruits
- Ca content of fruits
- firmness of apples
- total acid content of fruits
- quality index for fruits

### Results

The quality of the Jonathan apples is decisively influenced by the strength of the red skin colour which plays the greatest role in its external appearance. Figure 1 shows the 6-year average effect of the treatments on the skin colour of Jonathan apples. The two dolomite- and the limestone dust treatments equally significantly increased the proportion of fruits with more

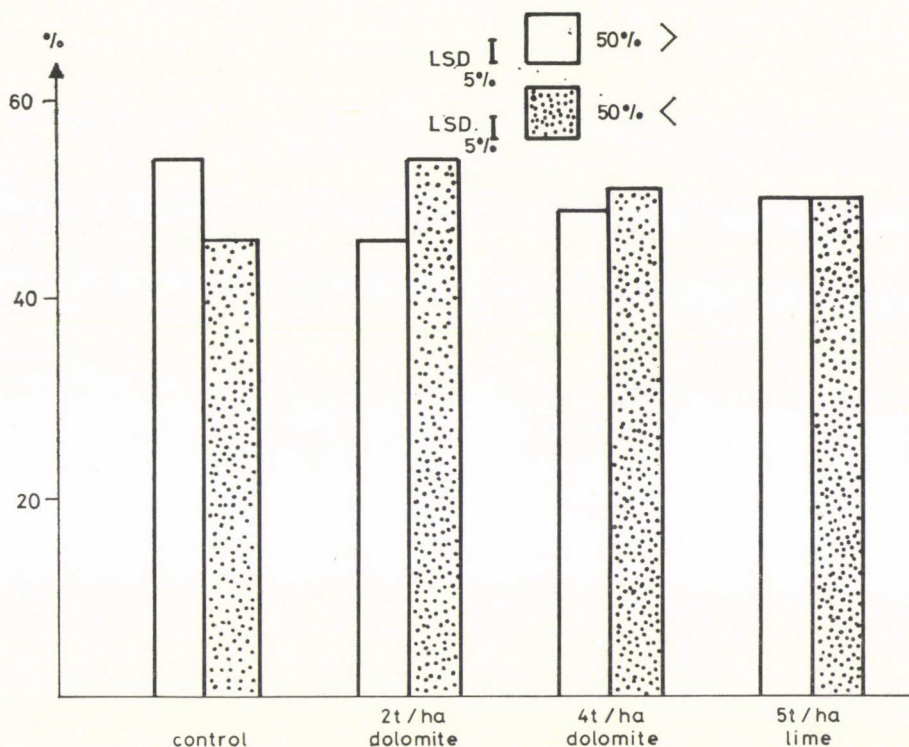


Fig. 1. Effect of dolomite- and limestone dust treatments on the size of the covering colour of apples

than 50% skin colour compared to the control. The red skin colour increased to the greatest extent in the case of 2 t/ha dolomite supplied, though both dolomite treatments considerably increased the size of the covering colour. This contradicts the statement of Kim-Lee (1980) who found that the dolomite treatments did not influence the colouring of the apples.

The firmness of the fruits of pomiferous plants is influenced by the weather and the agrotechnics as well as by the genetic characteristic of the variety. Lime- and dolomite fertilization through the soil usually did not directly increase the firmness of apples (Van der Boon 1966, Kim-Lee 1980): In Table 1. the effects of the treatments on the firmness of the apples, both when they were harvested and when they were removed from storage are shown. At the time of harvesting, the flesh firmness of the apples was found to have been increased by the dolomite treatments in three and by liming in four years of the experiment. On the average of the experimental years both dolomite treatments and the treatment with limestone dust resulted by the time of harvest in increased flesh firmness compared to the control. By the end of storage the flesh firmness of the apples of the dolomite- and



Table 1

*Effects of dolomite- and limestone dust treatments on the flesh firmness of Jonathan apples at the time of harvesting and at the end of storage ( $N/cm^2$ ) (1981–1986)*

	1981/82		1982/83		1983/84		1984/85		1985/86		5 year average		Percentage value of storing in
	B	E	B	E	B	E	B	E	B	E	B	E	
Control	75.3	46.8	77.7	49.0	60.1	57.8	76.6	49.6	88.9	52.1	75.7	51.1	67.5
Dolomite 2 t/ha	77.9	45.9	84.5**	44.4	66.0**	53.7	74.4	52.4	84.2*	55.0	77.4*	50.3	65.0
Dolomite 4 t/ha	79.4*	46.8	85.6***	48.1	64.3*	52.2	74.8	48.6	85.7	54.7	78.1	50.1	64.2
Limestone dust 5 t/ha	82.3***	49.9*	81.4	45.4	65.4*	51.5	76.0	49.2	92.6*	55.6	79.5	49.9	62.8
L. S. D. 5%	2.7	3.0	3.4	n.s.	n.s.	n.s.	n.s.	n.s.	3.6	n.s.			

Note:

B = beginning of storage

E = end of storage

\* — S. D. 5% difference

\*\* — S. D. 1% difference

\*\*\* — S. D. 0.1% difference

Storage period: 1981/82 171 day

1982/83 125 day

1983/84 166 day

1984/85 137 day

1985/86 154 day

Table 2

*Effects of dolomite- and limestone dust treatments on the total sugar content of Jonathan apples at the time of harvesting and at the end of storage (%) (1981–1986)*

	1981/82		1982/83		1983/84		1984/85		1985/86		5 year average		Percentage value of storing in
	B	E	B	E	B	E	B	E	B	E	B	E	
Control	10.6	10.3	12.2	12.7	11.7	11.0	11.4	10.2	10.3	10.0	11.2	10.8	96.4
Dolomite 2 t/ha	10.8	9.9	11.4*	11.0	12.2**	10.7	10.5*	10.0	10.3*	9.5	11.0*	10.2**	91.9
Dolomite 4 t/ha	12.2***	10.6	11.5*	11.3	12.6*	11.0	10.4*	10.6	10.5	10.0	11.5*	10.7	93.9
Limestone dust 5 t/ha	10.8	10.8	12.6	12.1	12.8*	11.4	11.3	9.8	10.5*	9.6	11.6***	10.7	92.2
L. S. D. 5%	0.3	0.8	0.6	n.s.	n.s.	0.6	0.6	0.5	0.5	n.s.	0.2	0.3	

Note:

B = beginning of storage  
 E = end of storage  
 \* — L. S. D. 5% difference  
 \*\* — L. S. D. 1% difference  
 \*\*\* — L. S. D. 0.1 % difference

Storage period: 1981/82 171 day  
 1982/83 125 day  
 1983/84 166 day  
 1984/85 137 day  
 1985/86 154 day



Table 3

*Effects of dolomite- and limestone dust treatments on the total acid contents of Jonathan apples at the time of harvesting and at the end of storage (1981–1986)*

	1981/82		1982/83		1983/84		1984/85		1985/86		5 year average		Percentage value of storing in
	B	E	B	E	B	E	B	E	B	E	B	E	
Control	0.71	0.44	0.63	0.46	0.65	0.39	0.93	0.55	0.66	0.60	0.72	0.49	73.1
Dolomite 2 t/ha	0.74	0.42	0.59*	0.55	0.63	0.39	0.76***	0.46***	0.65	0.47***	0.67***	0.46***	67.7
Dolomite 4 t/ha	0.74	0.37**	0.69**	0.55	0.69	0.40	0.91***	0.55	0.76***	0.53***	0.74*	0.48	64.9
Limestone dust 5 t/ha	0.78***	0.45	0.67*	0.59	0.68	0.41	0.84***	0.54	0.66	0.54***	0.73	0.51*	69.9
L. S. D. 5%	0.03	0.03	0.02	0.04	0.03	n.s.	0.04	0.02	0.03	0.02	0.02	0.01	

Note:

B = beginning of storage

E = end of storage

\* — L. S. D. 5% difference

\*\* — L. S. D. 1% difference

\*\*\* — L. S. D. 0.1% difference

Storage period: 1981/82 171 day

1982/83 125 day

1983/84 166 day

1984/85 137 day

1985/86 154 day

Table 4

*Effects of dolomite- and limestone dust treatments on the quality index of Jonathan apple at the time of harvesting and at the end of storage (1981-1986)*

	1981/82		1982/83		1983/84		1984/85		1985/86		5 year average		Percentage value of storing in
	B	E	B	E	B	E	B	E	B	E	B	E	
Control	176.7	146.0	185.0	173.1	182.0	148.7	206.3	157.0	169.0	160.0	183.9	157.0	87.4
Dolomite 2 t/ha	182.3	141.0	173.0	164.7	186.0	146.0	181.7	146.0**	168.0***	142.0	178.2	147.9	83.0
Dolomite 4 t/ha	196.3***	143.7	184.0	168.0	194.0***	151.0	185.7	161.7	181.0***	153.3*	188.3***	155.0	82.5
Limestone dust 5 t/ha	186.7**	152.3	193.9*	180.3	196.6***	155.3	196.7	152.7	171.7	150.7*	188.9***	158.3	83.6
L. S. D. 5	5.9	8.2	6.6	n.s.	n.s.	5.7	5.7	6.3	2.9	n.s.			

Note:

B = beginning of storage  
 E = end of storage  
 \* - L. S. D. 5% difference  
 \*\* - L. S. D. 1% difference  
 \*\*\* - L. S. D. 0.1% difference

Storage period: 1981/82 171 day  
 1982/83 125 day  
 1983/84 166 day  
 1984/85 137 day  
 1985/86 154 day



Table 5

*Correlation between the Mg content of the 0–60 cm layer of soil and some indices of the leaf and fruit in the apple variety Jonathan on the average of 6 years (1980–1985)*

Factors	Correlation coefficient <i>r</i>	Number of elements <i>n</i>
1. Mg content of leaf (%)	–0.189	28
2. Ca content of leaf	–0.302	28
3. Firmness of fruit	0.071	72
4. Acid content of fruit	–0.007	72
5. Quality index for fruit	0.097	72
6. Mg content of fruit (in term of fresh weight)	0.569***	72
7. Ca content of fruit (in term of fresh weight)	0.733***	72

Note: \*\*\* The correlation is significant at L. S. D. 0.1%

lime treatments slightly decreased compared to the control, on the average of the experimental years.

The effects of the dolomite- and limestone dust treatments on the total sugar contents of the apples are seen in Table 2. No definite, tendency-like effect of treatment concerning the total sugar content could be observed either at the time of harvesting or at the end of storage. The effects compared to the control are uncertain and vary from year to year.

The 4 t/ha dolomite treatment and liming resulted in a significantly increased total acid content at the time of harvesting in three out of the five years of the experiment; so these effects were also significant on the average of the experimental years (Table 3).

In the 2 t/ha dolomite treatment the apples contained significantly less acid in the last two years and on the average of the experiment. In the dolomite- and lime fertilization treatments, the total acid content expressed as a percentage of the value obtained at the time of harvesting decreased to a greater extent compared to the control.

The quality index calculated by the method of Thiault (1970) was significantly higher at the time of harvesting, with the 4 t/ha dolomite treatment in three, with the lime treatment in two out of the five experimental years. With these two treatments significantly higher quality index values were also obtained on the average of the five experimental years (Table 4).

In Table 5 the major results of the examination of correlations between the magnesium content in the 0–60 cm soil layer and some factors considered are given on the average of the years 1980–1985. No significant close correlation was found between the magnesium content of the 0–60 cm layer of the soil and the Mg- and Ca contents of the leaves and acid content, flesh

firmness and quality index of the fruits, respectively, a close significant correlation was observed on the other hand, between the magnesium content of the soil and the Mg- and Ca contents of the apples.

### References

- Allen, M. (1980): *Relationships between soil factors and magnesium deficiency in apple*. (In: Atkinson, D., Jackson, J. E., Sharples, R. O., Waller, W. M.: Mineral nutrition of fruit trees, Butterworths, London-Boston), 279-283.
- Balogh, I., Blaskó, L., Nyiri, L. (1984): Research results on the liming of acid soils and the joint application of Ca and Mg. *CIEC 9<sup>th</sup> World Fertilizer Congress Proceedings*, 3, 35-39.
- Bergmann, W. (1983): *Ernährungsstörungen bei Kulturpflanzen*. VEB Gustav Fischer Verlag, Jena.
- Boynton, D., Oberly, G. H. (1966): *Apple nutrition*. (In: Childers, N. F.: Nutrition of Fruit Crops), Rutgers, New Brunswick, 1-50.
- Dione, J. L., Roy, P. O., Allard, O., Granger, R. L. (1967): The effect of nitrogen, potassium and magnesium on two McIntosh apple orchards. *Can. J. Plant Sci.* 47, 563-570.
- Fisher, E. G., Walker, D. R., Boyton, D., Kwoug, S. S. (1958): Studies on the control of magnesium deficiency and its effect on apple trees. *Proc. Amer. Soc. Hort. Sci.* 71, 1-10.
- Ford, E. M. (1964): The control of magnesium deficiency in apple rootstock stoolbeds. *J. Hort. Sci.* 39, 212-223.
- Hansen, P. (1975): Virking af kalium og magnesium til "Cox's Orange" på lermuld. *Tidsskr. Planteavl.* 79 (2), 259-265.
- Kim, K. R., Lee, Y. C. (1980): Effects of dolomite and epsom salts application on apple trees. 21 (2), 164-169.
- Lord, W. J. (1983): Nutritional problems in 1982 and suggestions for fertilization of apple trees in 1983. *Fruit Notes* 48 (2), 1-5.
- Papp, J. (1988): Improvement in the magnesium supply of apple orchards using dolomite. *Acta Agronomica Hungarica*, 37, (3-4), 191-195.
- Pedersen, B. F., Vang-Petersen, O. (1984): Magnesium til æble. II. *Fidsskr. Planteavl.* 88, 405-412.
- Sadowski, A., Jadczyk, E., Pekacki, J., Scibisz, K. (1976a): Magnesium nutrition of apple trees. II. Effect of K and Mg fertilization and comparison of soil and foliar treatments with magnesium sulphate. *Acta Agrobotanica*, 29 (2), 179-199.
- Sadowski, A., Jadczyk, E., Poltorak, M., Scibisz, K. (1976b): Magnesium nutrition of apple trees III. Comparison of different methods of magnesium fertilization. *Acta Agrobotanica*, 19 (2), 201-217.
- Thiault, J. (1970): Etude de critères objectives de la qualité gustative de pomme Golden Delicious. *Bull. Techn. d'Information*. 248, 1-11.
- Trocme, S. (1960): La magnésie en arboriculture fruitière. *Arboriculture Fruitière*, 7 (82), 10-13.
- Van der Boon, J., Das, A., Schreven, A. C. (1966): A five year fertilizer trial with apples on sandy soil, the effect on magnesium deficiency, foliage and fruit composition and keeping quality. *Neth. J. Agr. Sci.* 14, 1-31.
- Whitear, J. D. (1978): *The magnesium way to healthy crops*. Feature, London Press Service, No. 1019/8.





## THE EFFECT OF DIFFERENT N-DOSES ON CHANGES IN THE NITRATE-SUGAR AND CAROTENE CONTENTS OF CARROT

I. CSERNI, K. PROHÁSZKA and I. PATÓCS

VEGETABLE RESEARCH INSTITUTE DEVELOPMENT ENTERPRISE, KECSKEMÉT, HUNGARY

(Received: 29 October 1987; accepted in revised form: 15 February 1988)

Under Hungarian ecological conditions during the vegetation period of carrot, twice the amount of precipitation should be supplemented by irrigation in years of drought; while under average precipitation, the amount of irrigation water should be so increased as to redouble it.

At the beginning of the vegetation period the  $N_{160}$  kg/ha treatment (with a  $P_{140}$  and  $K_{360}$  kg/ha basic fertilization of  $P_2O_5$  and  $K_2O$  as active agents) proved the best. The average samples taken at the beginning of September gave the largest shoulder breadth and weight and the foliage was healthy and uniformly green throughout the vegetation in this treatment. In response to higher rates of nitrogen application, the leaves turned dark green as a sign of overdosing. In the treatments given less than 160 kg/ha nitrogen the pale green to light yellowish green colour of leaves revealed a nitrogen deficiency.

For this very reason the quantity of N active agent ( $N_{320}$  kg/ha) calculated on the basis of the nutrient balance is considered to be too large; it may be uneconomical and may even result in environment pollution.

According to the analysis of components, N fertilization seems to influence the carotene content positively.

The 320 kg quantity of N calculated after the nutrient balance increased the  $NO_3$ -content of the carrot roots 3-4-fold compared to the lower rate treatments. The provocative dose of  $N_{640}$  kg/ha raised the nitrate level far above the permissible level (400 ppm).

Under irrigated conditions, smaller quantities of nitrogen divided in three parts are easily washed out from sandy soils and thus do not cause a harmful increase in the  $NO_3$  content of the roots.

Increased rates of N-fertilization, while having a negative influence on the quantity of dry matter, exercised a positive effect on the carotene content.

**Keywords:** carotenes, carrot, *Daucus carota* L., nitrates, N-fertilization

### Introduction

The  $NO_3$  content of vegetable crops grown for fresh consumption can be influenced by many factors.

Analyses of nitrate contents are made necessary by the harmful effect of larger quantities of  $NO_3$  on the human organism in general, and particularly that of infants. The quantity of  $NO_3$  contained in dietetic baby-food is an even more serious question: it must not exceed 50 ppm. In countries outside Hungary for each vegetable used as basic material in producing baby-food, the maximum permissible  $NO_3$  content is prescribed by standard.



According to the Hungarian standard, the maximum permissible  $\text{NO}_3$  content of baby-foods in 400 ppm (decree 4/1978. (VI. 25.) EüM, while e.g. in the German Democratic Republic it is 300 ppm.

According to the results of our earlier small culture pot experiments with carrot, in the course of the vegetation period the  $\text{NO}_3$  content decreased irrespective of the nutrient treatments.

A comparison of varieties showed the long vegetation variety "Fertődi vörös" to have the lowest  $\text{NO}_3$  content (156 ppm), while the "French" varieties of shorter vegetation gave a higher (270 ppm) average value (Cserni et al. 1983).

### Materials and methods

The experiment was set up in large (250 l) culture pots sunk in the soil, on a sandsoil of Kecskemét, arranged at random with 4 replications of each of 6 treatments, on an area of  $0.30 \text{ m}^2$ . The humus content of the experimental soil was very poor, its AL-P and AL-K content was medium.

Fertilization for treatment 5 was determined on the basis of soil analyses, with a 60 t/ha yield in view, and the nutrient status of the soil and the nutrient demand of the carrot taken into consideration.

The rate of P- and K-fertilization was left unchanged while the amount of N-fertilizer was gradually decreased or increased (Table 1).

The basic fertilizers (P and K) were mixed with the previously homogenized soil to a depth of 25 cm prior to setting up the experiment. The amount of N was divided in three equal parts; one-third was spread over the surface immediately after sowing (on 23 April 1986, in the form of 34% ammonium nitrate), one-third was given on 2 June 1986, and the final-third on 2 July 1986.

Phenological observations were repeatedly made during the vegetation period and the results were included in a table.

In the experiment, dripping irrigation was applied; the amount of water supplied per unit area is given in mm.

Besides the  $\text{NO}_3$  content, the values of dry matter-, simple- and compound sugar- and carotene contents were also determined for the carrot variety "Fertődi vörös", as a function of N-supply.

The first samples taken at random (10 plants/pot) were individually examined for root length, shoulder width and weight on 2 September 1986. Afterward each root was cut into four lengthwise; two quarters were analysed for components, while the remaining two quarters were frozen to determine their  $\text{NO}_3$  content for second time. The  $\text{NO}_3$  content was first de-

**Table 1**  
*Treatments in the experiment*  
(1986)

Treatments	N	$\text{P}_2\text{O}_5$		$\text{K}_2\text{O}$
	kg/ha	active	agent	
1	0	140		360
2	40	140		360
3	80	140		360
4	160	140		360
5	320	140		360
6	640	140		360

Table 2

*Quantities of precipitation and irrigation water during the vegetation period  
(Culture pot experiment with carrot, 1986)*

Vegetation period	50 years' average precipitation mm	1986		
		Precipitation mm	Irrigation water mm	Precipitation + irrigation water mm
April	44.0	29.5	—	29.5
May	55.0	27.4	41.7	69.1
June	54.0	88.1	62.5	150.9
July	47.0	51.9	138.9	190.8
August	44.0	26.5	166.7	193.2
September	45.0	0.4	69.4	69.8
October	47.0	0.0	41.7	41.7
Total	336.0	224.1	520.9	745.0

terminated immediately after lifting the roots; therefore only 6 samples (1 series) were examined a day.

For determining the  $\text{NO}_3$  content 20 g of the carrot, rasped down and homogenized, was reduced to pulp, put into 100 ml of 1%  $\text{CuSO}_4$  solution, then kept in water-bath for 30 minutes. The solution was then filtered, and the  $\text{NO}_3$  content of the filtrate measured by an OP-211/1 pH-meter with OP- $\text{NO}_3$ -0711 P selective  $\text{NO}_3$ -sensitive electrode against a 08-0820 P reference electrode (Binder et al. 1984).

The carotene content was determined after Walger and Thuranszky (1962); the simple sugar with Fehling reaction, and so also was the compound sugar after inversion.

## Results

### *Meteorological data*

The quantitative data of precipitation and irrigation water are contained in Table 2.

As seen from the data, the amount of precipitation during the vegetation period was 224.1 mm, about 100 mm less than the 50-years average. During the growth season an amount of irrigation water equal to 520.9 mm precipitation was supplied, which together with the precipitation (745 mm) resulted in favourable, uniform development.

Under the ecological conditions of Hungary in dry years such as this, twice the amount of rainwater should be supplied in the form of irrigation water, but even under normal precipitation conditions the amount of irrigation water should be so increased as to redouble the precipitation water, if a uniform development is to be ensured.

### *Observations during the vegetation period*

The results of phenological observations during the vegetation period are summarized in Table 3 on the basis of scores from 1 to 5, and colour, evaluation.



Table 3

Qualitification of the carrot variety "*Fertődi vörös*" during the vegetation period (1986)  
(On the basis of scores from 1 to 5 and the colour of leaf)

Treatment	2 June 1986	18 June 1986	15 July 1986
1	3.1	1.9 pale green	1.0 pale greenish yellow
2	3.5	2.0 slightly light green	2.3 light greenish yellow
3	3.5	3.1 light green	3.5 greenish yellow
4	4.3	4.3 green	4.5 green
5	3.5	5.0 dark green	5.0 dark green
6	3.0	4.6 fierce green	5.0 fierce green

Treatment 1 ( $N_0$ ) hardly differed in colour from the other treatments at the beginning of the vegetation period. With the advance of vegetation a nitrogen deficiency became more and more visible: from light green the leaves turned into pale yellow.

Treatment 2 ( $N_{40}$ ) showed at the beginning imperceptible colour differences and negligible differences in development, compared to the other treatments. On the second occasion of qualification, there was hardly any difference in development compared to the control; the light green colour of the leaves was slightly different from the colour of the control. In mid-July the differences both in development and colour were substantial compared to the control, indicating again a nitrogen deficiency.

Treatment 3 ( $N_{80}$ ) did not show any difference in development at the beginning. Later both the foliage and the root were of medium development. In the course of the vegetation period the leaves became light green or greenish yellow.

Treatment 4 ( $N_{160}$ ) was the best of all at the beginning of the vegetation period; the plants remained healthy, fine and uniformly green to the end.

Treatment 5 ( $N_{320}$ ) gave a relatively poor stand at the beginning while later the foliage became thick and dark green as a sign of nitrogen overdosing.

In treatment 6 the provocative rate of  $N_{640}$  fertilization had a depressive effect. The strong green colour of the leaves undeniably indicated the over-application of nitrogen.

### Results of root measuring

In Table 4 the average length, shoulder width and weight of the carrot roots examined are given.

As seen from the results, the longest roots were obtained with the  $N_{320}$  treatment. The shoulder width and root weight were the same in Treatments

Table 4

*Average root length, shoulder width and root weight of the carrot variety  
"Fertődi vörös" Kecskemét, 1986*

Treatments	R o o t					
	Length		Shoulder width		Weight	
	2 Sept.	23 Oct.	2 Sept.	23 Oct.	2 Sept.	23 Oct.
1	16.1	125	2.8	28	38	37
2	17.4	128	3.2	29	60	40
3	17.5	135	3.1	30	54	45
4	18.7	148	3.4	31	71	49
5	19.3	154	3.2	30	64	56
6	18.0	122	3.2	27	64	45
L.S.D. 5%		12		2		9

N<sub>320</sub> and N<sub>640</sub>. From Treatment 1 to 5 the length of root gradually increased. The width of shoulders was the smallest in Treatment 0.

The shoulder-width and weight of roots gave the highest values in the N<sub>160</sub> treatment which also showed a uniformly developed fine green stand throughout the growth season.

#### *Analysis results of components*

According to the results of carrot analyses there were no significant differences in dry matter-, simple- and compound sugar content in response to nitrogen fertilization (Table 5).

Table 5

*Dry matter-, simple and compound sugar-, carotene- and NO<sub>3</sub> contents of the  
carrot variety "Fertődi vörös" as a function of N treatments  
Kecskemét, 1986*

Treat- ments	Dry matter %		Simple sugar %		Total sugar %		Carotene mg/100 g dry matter		NO <sub>3</sub> ppm	
	2 Sept.	23 Oct.	2 Sept.	23 Oct.	2 Sept.	23 Oct.	2 Sept.	23 Oct.	2 Sept.	23 Oct.
1	13.29	13.17	0.49	0.88	5.36	5.67	88.54	98.42	53.00	66.75
2	13.26	13.95	0.66	0.75	5.64	5.92	94.20	96.60	63.25	71.50
3	12.36	13.42	0.66	0.84	5.26	5.73	108.03	100.30	53.00	68.00
4	12.56	13.80	0.85	0.77	5.06	5.93	116.34	108.90	69.75	77.00
5	12.35	13.41	0.68	1.19	4.65	5.93	117.28	120.67	192.50	115.75
6	12.21	15.17	0.57	1.10	4.48	6.34	119.14	98.89	915.25	362.25
L.S.D. 5%	—	0.94	—	0.31	—	0.52	15.14	14.64	270.00	158.73



Table 6

Correlation matrix for the N-dose and the dry matter-, simple and compound sugar-, carotene- and  $\text{NO}_3$  contents of the carrot variety "Fertődi vörös" ((2-5 September 1986))

		N-dose	Dry matter	Simple sugar	Compound sugar	Carotene	$\text{NO}_3$
		1	2	3	4	5	6
N-dose	1	1.0000	-0.3028	-0.0219	-0.5105	0.4880	0.8305
Dry matter	2		1.0000	-0.1980	0.3934	-0.1726	-0.1905
Simple sugar	3			1.0000	0.2045	0.2427	-0.0702
Compound sugar	4				1.0000	0.2251	-0.3596
Carotene	5					1.0000	0.1891
$\text{NO}_3$	6						1.0000

As for the carotene content differences significant at 5% level were found in the  $\text{N}_{160}$ ,  $\text{N}_{320}$  and  $\text{N}_{640}$  treatments compared to the  $\emptyset$  and low rate ( $\text{N}_{40}$ ) treatments.

The carotene content increased in direct ratio to the increase in the rate of N application.

Lower ratios of N fertilization caused no differences in  $\text{NO}_3$  content, while in the  $\text{N}_{320}$  treatment it increased about 3-4-fold. The provocative rate of  $\text{N}_{640}$  caused a significant increase in the  $\text{NO}_3$  content of the root compared to all the other treatments.

The results of  $\text{NO}_3$  determination seem to prove that only an extremely high rate of nitrogen fertilization ( $640/3 = 213 \text{ kg/ha}$  N active agent) applied in the last stage of root development increases deleteriously the nitrate content of the carrot root. The  $160/3 = 53 \text{ kg/ha}$  quantity of nitrogen, on the other hand, did not practically increase the  $\text{NO}_3$  content.

Correlation matrix calculations indicated a medium negative correlation between the rate of N fertilization and the dry matter content. The correlation was found to be medium between the applied quantity of N and the carotene content, and close between the rate of N and the  $\text{NO}_3$  content.

The dry matter content showed a medium close correlation with the quantity of compound sugar.

## Discussion

Too high  $\text{NO}_3$  contents occur primarily in forced vegetables. Literary data and our own results testify that the  $\text{NO}_3$  content of vegetable crops may greatly vary according to species and variety (Claus 1983, Cserni et al. 1983, Derolez-Vulsteke 1985, Terbe-Cserni 1986, Terbe et al. 1986 etc.). Furthermore, the quantity of  $\text{NO}_3$  is greatly influenced by environmental



factors. So the  $\text{NO}_3$  content depends on the light conditions, temperature, age and size of the plant, nutrition — more exactly on the amount of N-fertilizer, that is on the nutrient status of the soil. It also depends on the water content of the soil, the plant density, the length of time from harvesting to storage and to use, respectively, and possibly on hitherto unknown factors. Under glasshouse conditions the  $\text{NO}_3$  content can be 2–3-times as high as in the field.

Scharp et al. (1986) reduced the nitrogen fertilizer active agent from 240 to 110 kg/ha whereby the  $\text{NO}_3$  content of the soil also decreased. Brückner (1986) too recommends an about 100 kg N active agent quantity as sufficient for a large yield and preventing the  $\text{NO}_3$  content from rising high. The high  $\text{NO}_3$  content of the soil and an excessive  $\text{NO}_3$  uptake of plants as a consequence can be decreased by a reasonable succession of crops as well.

According to Derolez (1985), in agreement with our own investigations (Cserni et al. 1983) the  $\text{NO}_3$  content in carrot depends on the age and size of the plant. The smaller and younger the carrot, the higher its  $\text{NO}_3$  content.

Polach (1982) is of the opinion that the  $\text{NO}_3$  content depends on the cropyear (it is probably affected by insufficient light conditions and low temperatures).

As known from recent literary data, in Holland the maximum  $\text{NO}_3$  content of lettuce is fixed at 4500 ppm from November to March, and at 3500 ppm subsequently (Schaer and Habben 1986). In Switzerland this value is 3500 ppm regardless of the season.

Of the environmental factors which act on the  $\text{NO}_3$  content, only the nutrient status and -ratio of the soil can be substantially influenced, and that by choosing the optimum time of the last nitrogen top-dressing.

Basic fertilization and N top-dressing with the view of keeping the  $\text{NO}_3$  content of plants at an acceptable level and obtaining optimum yield per  $\text{m}^2$  can only be reasonably carried out on the basis of soil analyses. In addition, repeated probative  $\text{NO}_3$  measuring will be necessary during the vegetation period in order to determine the optimum rate and time of N top-dressing, depending on species, variety, soil type, nutrient status and harvesting time, because the accumulation of  $\text{NO}_3$  in the plant is influenced not only by the amount of nitrogen contained in the soil and supplied by fertilization but also by the ratio of other nutritive substances (N, P, K, Ca, Mg) in the soil.

### References

- Binder, I., Czecei, L., Schlotter, Gy. (1984): Nitráttartalom vizsgálatok összehasonlító értékelése (Comparative evaluation of nitrate content analyses). *Hűtőipar*, **30** (2), 50–54.  
 Brückner, U. (1986): Nährstoffversorgung von Möhren. *Gemüse*. **22** (2), 58–60.  
 Claus, P. (1983): Nitrat im Gemüsebau — ein Umwelt- und Qualitätsproblem. *Deutscher Gartenbau*. **3**, 1371–1374.



- Cserni, I., Prohászka, K., Vidéki, L. (1983): A sárgarépa tápanyaggazdálkodásának tanulmányozása tenyészedeny-kísérletben (Nutrient regime studies in carrots grown in containers). *Zöldségtermesztési Kutató Intézet Bulletinje*. Kecskemét. **16**, 95–107.
- Klein, E. (1980): *Vergleich gärtnerischer Produktions-systeme in Rahmen des Forschungsvorhabens "Gesunde Ernährung"*. Jahresbericht 1979/80. Bayerische Landesanstalt. Würzburg. 86–88.
- Polách, J. (1982): Vliv hrojeni na vynos a kvalitu mrkve (nitrátovydusik). *Bull. vyzk. a slecht. Ust. Zelin. Olomouc*. 25/26. 1981–1982. 119–127.
- Robio, R. (1982): Les nitrates dans les laitues pommées. *Revue Horticole Suisse*. **10**, 295–298.
- Schaez, T., Habben, J. (1986): Nitratgehalt von Kopfsalatsorten unter Glas. *Gemüse*. **22** (3), 104–105.
- Scharp, H. C., Budig, M., Wehrmann, J. (1986): Das Nitratproblem — Wege zur Lösung. *Gemüse*. **22** (3), 113–115.
- Terbe, I., Cserni, I. (1986): A zöldségnövények  $\text{NO}_3$ -tartalmának és az azt befolyásoló tényezőknek a vizsgálata. Kecskemét. 1–10. p. (manuscript).
- Terbe, I., Patócs, I., Zsoldos, L. (1986): Zöldségnövények nitráttartalma (Nitrate content of vegetable crops). *Hajtatás, Kofai Termesztés*. **18** (12), 10–12.
- Walger, J., Thuránszky, A. (1962): Egyszerű módszer zöldségnövények és szénák karotintartalmának meghatározására (A simple method to determine the carotene content of green plants and hay). *Agrokémia és Talajtan*. **11** (3/4), 443–454.

## THE EFFECT OF PLASTIC TUNNEL ORIENTATION ON YIELD OF SOME CUCUMBER VARIETIES IN EGYPT

FAROUK EL-AIDY

FACULTY OF AGRICULTURE, KAFR EL-SHEIKH, EGYPT

(Received: 9 September 1987; accepted in revised form 14 December 1987)

Eighteen cucumber varieties were cultivated under plastic tunnels erected in two orientations North/South and East/West, during the winter season of 1984/1985.

Data reveal that the N/S orientation led to the production of the higher total yield, while the E/W orientation improved the early yield.

Cucumber varieties differed relatively in their response to the orientation of the tunnel. N/S orientation was more suitable for long fruited varieties, while E/W orientation was more suitable for short fruited varieties in the Egyptian climate.

**Keywords:** cucumber, *Cucumis sativus* L., plastic tunnel, orientation, yield

### Introduction

Egypt has an arid mediterranean climate. The temperature becomes too low in winter and too high in summer for favourable cucumber production. Recently this has been overcome by introducing protected cultivation which raises the temperature in winter. This has increased the cucumber yield (El-Aidy and Moustafa, 1977).

During last year the plastic tunnel technique has been used commercially in vegetable crop production in Egypt (about 2000 plastic houses each about 500 m<sup>2</sup>; more than 90% of them were cultivated by cucumber).

Although Egypt receives a lot of winter sunlight the orientation and the form of greenhouses are very important because of the shadow produced by frame. In Portugal it was found that the highest inside radiation, occurred under a single span greenhouse orientated E/W with roof slope of about 20 degrees (Monteiro, 1984).

The value of this study is important for Egypt, and other countries having the same climate, since they mainly use the unheated plastic tunnels during winter. Any improvement in the use of such tunnels would lead to the improvement of crop production.



## Materials and methods

Experiments were carried out at Kafr El-Sheikh, Faculty of Agriculture Farm during the 1984–1985 winter season. Two tunnels, each 7.5 m wide, 3 m high and 24 long clad by clear PE film (0.15 mm thick) were used. One of them was oriented to E/W and the other to N/S.

Eighteen cucumber varieties were planted in November 1984, at a density of 4 plants/m<sup>2</sup>. First pick was recorded on 15 February 1985. The experiment was completed at the end of April.

A completely randomized block design with four replicates — each 2.5 m<sup>2</sup> was adopted. Duncan's multiple range test was used for the comparisons among the treatment means (Duncan, 1965).

Insect and disease control programs and other cultural methods were done as practiced by local farmers.

## Discussion

Data in Table 1 show the average yield of the eighteen cucumber varieties cultivated under plastic tunnels. The results indicate that there is a big variation among the varieties under the same conditions, despite the fact that these varieties are in good character.

The best results were obtained from "Farbiola" (11.0 kg/m<sup>2</sup>) and "Corona" (9.6 kg/m<sup>2</sup>) which are long fruited types and "Biet Alpha K. 2744"

Table 1

*Average yield of different cucumber varieties cultivated under plastic tunnels during winter season*

Variety	Kg/plot	Kg/m <sup>2</sup>
Farbiola	27.5	11.0
Corona	24.0	9.6
Biet Alpha K.2744	23.7	9.5
K. 1700	23.2	9.3
Dalibor	22.3	8.9
Picobello	22.1	8.8
T. W. 871	21.4	8.5
Biet Alpha H <sub>1</sub>	20.7	8.3
Avir	20.5	8.2
Bambola	18.0	7.2
Arabio	17.7	7.1
Pepinova	16.6	6.6
Fidelio Improved	15.6	6.2
Super Mini Esmaralda	12.8	5.1
Carabel	12.6	5.0
Profito	12.3	4.9
Donor	11.1	4.4
Biet Alpha	11.0	4.4
L.S.D. <sub>5%</sub>	2.9	—

(9.5 kg/m<sup>2</sup>) and "K. 1700" (9.3 kg/m<sup>2</sup>) from short fruited types. Although these results are less than those obtained in Holland (4), the production in Egypt can be considered of great importance when compared to open field production which amounted to 1.25 kg/m<sup>2</sup> in the summer and 0.40 kg/m<sup>2</sup> in the winter (3).

The results shown in Table 2 indicate the effect of orientation of plastic tunnel on the total yield of cucumber cultivated during the winter season. The data demonstrate that there are significant differences between N/S

Table 2

*Effect of orientation of plastic tunnel on the total yield of cucumber*

Orientation	Kg/plot
N/S	20.75
E/W	16.68
L.S.D. <sub>.5%</sub>	0.768
L.S.D. <sub>.1%</sub>	1.020

Table 3

*Effect of orientation on total yield of cucumber varieties (kg/plot)*

Varieties	N/S	E/W	F.test
Farbiola	29.3	25.6	*
Corona	30.2	17.8	**
Biet Alpha K.2744	22.4	25.0	N.S.
K. 1700	22.0	24.4	N.S.
Dalibor	27.2	17.4	**
Picobello	21.2	23.0	N.S.
T. W. 871	25.2	17.0	**
Biet Alpha H <sub>1</sub>	20.4	21.0	N.S.
Avir	22.4	18.6	*
Bambola	22.9	13.2	**
Arabio	21.1	14.2	**
Pepinova	20.3	12.8	**
Fidelio Improved	16.3	14.8	N.S.
Super Mini Esmaralda	15.0	10.6	*
Carabel	14.2	10.9	*
Profito	13.0	11.6	N.S.
Donor	13.7	8.6	**
Biet Alpha	12.4	9.6	N.S.
L.S.D. <sub>.5%</sub>	3.44		*
L.S.D. <sub>.1%</sub>	4.57		*



and E/W orientation. The difference was 1.7 kg/m<sup>2</sup>. The highest average yield was obtained from N/S orientation. These results indicate that the ventilation, even during the winter period in Egypt, is more important than the heating, because the N/S position produces better ventilation, while E/W position gives higher solar radiation (5).

The interaction between the effect of orientation and the cucumber varieties is shown in Table 3. The results indicate a great difference between the response of cucumber varieties to the orientation of the plastic tunnel. Most of long fruited varieties have significant differences while the short fruited varieties were insignificant.

These results may be due to the more vigorous vegetative growth of those long fruited varieties which need more ventilation than those of short fruited varieties which have less vigorous growth.

### Conclusion

The results demonstrate that an excellent out-of-season crop of cucumber can be grown under unheated plastic tunnels in Egypt.

Cucumber varieties differ in their response to the orientation of the tunnels.

N/S orientation of the tunnel was more suitable for long fruited varieties, while E/W orientation was more suitable for short fruited varieties under Egyptian climate conditions.

### References

- Dancun, B. D. (1965): Multiple range and multiple F. tests. *Biometrics*, **11**, 1-42.
- El-Aidy, F., Moustafa, S. (1977): Cultivation of some cucumber varieties under plastic tunnels during winter. *J. Agric. Res. Tanta Univ.*, **3** (2), 157-161.
- El-Aidy, F. (1984): Research on the use of plastics and shade nets on the production of some vegetable crops in Egypt. *Plasticulture*, **61**, 55-59.
- Kwantitatieve Informatie Voor de Glastuinbouw, Proefstation voor Tuinbouw onder Glas, Naaldwijk, 1984-1985, G36-G55.
- Monteiro, A. (1984): In Portugal, protected cultivation of vegetables in the Algarve. *Plasticulture*, **64**, 9-16.

## EFFECT OF Fe AND Mn APPLICATION ON THE YIELD AND THEIR CONTENT IN BERSEEM (*TRIFOLIUM ALEXANDRINUM*) GROWN IN AN ALKALINE SOIL

R. L. BANSAL, D. S. CHAHAL and P. N. TAKKAR

DEPARTMENT OF SOILS, PUNJAB AGRICULTURAL UNIVERSITY  
LUDHIANA, INDIA

(Received: 21 September 1987; accepted in revised form 19 January 1988)

The effect of Fe and Mn application on the dry matter yield of berseem and their availability in an alkaline soil was studied in a greenhouse experiment. There were four levels of Fe and Mn; 0, 50, 100 and 200 mg kg<sup>-1</sup> soil. The dry matter yield increased significantly with the application of Fe or Mn at 50 mg kg<sup>-1</sup> soil, but at higher rates it decreased. The higher rates of applied Fe had more intense effect in decreasing the yield than the applied Mn. Fe application decreased the plant Mn and also the soil pH. The recovery of added Fe and Mn by DTPA extractant after the crop harvest was found to be 6.5% and 12.4% respectively.

**Keywords:** berseem, Fe-application, Mn-application, *Trifolium alexandrinum* L., yield

### Introduction

Manganese deficiency has been observed in berseem crops grown for more than five years on coarse textured highly permeable soils in rotation with rice (Takkar and Nayyar 1981). The deficiency has resulted from leaching losses of Mn and account of marked solubility during the period of soil submergence for rice. Under such conditions the available Fe also increases and the nutrient may have further depressed the absorption of Mn, and thereby contributed to Mn deficiency in berseem. Sideris and Young (1949) noticed that the high levels of Mn supply reduced the Fe concentration in the tissue of *Ananus comosus*. Nevertheless, Swarup and Mishra (1972) found a synergistic effect of Fe on Mn uptake in oats. Van der Vorm and Van Diest (1979) did not support the assumption that high levels of available Fe may have a suppressive effect on the uptake of Mn, and a high level of available Mn does not appear to have an adverse effect on the uptake of Fe by rice. Thus there are conflicting reports on the nature of the effect of Fe and Mn in plants. Since the information on this aspect is lacking, the present investigation therefore was undertaken.



## Materials and methods

A greenhouse experiment was conducted using a Fatehpur loamy sand (Ustipsamments) containing 82.0% sand ( $> 50 \mu$ ), 11.0% silt ( $2-50 \mu$ ), and 6.6 % clay ( $< 2\mu$ ). The soil had pH 8.9 and electrical conductivity  $0.3 \text{ mmhos} \cdot \text{m}^{-1}$  in 1 : 2 soil water suspension. The content of calcium carbonate and organic carbon were 0.80 and 0.24%. Available P and K (1 N ammonium acetate pH 7.0 extractable) were 12 and  $187 \text{ kg ha}^{-1}$ , respectively. The DTPA extractable (Lindsay and Norvell, 1978) Fe and Mn were 3.9 and  $1.6 \text{ mg kg}^{-1}$  soil. Polythene lined earthen pots were filled with three kg soil. The treatment comprised four levels each of Fe and Mn (0, 50, 100 and  $200 \text{ mg kg}^{-1}$ ) applied as  $\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$  and  $\text{MnSO}_4 \cdot \text{H}_2\text{O}$ . All the pots received basal application of 10 mg N and 30 mg  $\text{P}_2\text{O}_5 \text{ kg}^{-1}$  soil as urea and  $\text{KH}_2\text{PO}_4$ , respectively. These were repeated three times in a completely randomized design. The required quantity of salts were applied as solution to 3 kg soil and thoroughly mixed before filling the pots. Berseem was grown as a test crop. The crop was harvested after 60 days of germination. The representative soil samples were drawn from each pot after the harvest of crop.

The plant samples were washed successfully with 0.1 N HCl, distilled and deionized water, oven dried, weighed and ground in a Wiley Mill to pass a 40 mesh stainless steel screen. The plant samples were wet ashed with nitric-perchloric-sulphuric acid mixture. The content of Fe and Mn in the plant and soil extracts were measured by atomic absorption spectrophotometry.

## Results and discussion

### *Dry matter yield and uptake of Fe and Mn*

The dry matter yield of berseem crop increased significantly with the application of  $\text{Fe}_{50}$  or  $\text{Mn}_{50}$ , and beyond which it decreased (Table 1). The decline was more marked with the increasing rates of applied Fe (45% at  $\text{Fe}_{200}$ ) as compared to that of Mn (8% at  $\text{Mn}_{200}$ ). The response to Fe and Mn application indicated the deficiency of both the nutrients in the selected experimental soil. Gupta (1972) reported 50 per cent reduction in barley yield with the application of  $400 \text{ mg Fe kg}^{-1}$  soil.

Table 1  
*Effect of iron and manganese application on the dry matter yield and their content in berseem\**

Treatment $\text{mg kg}^{-1}$ soil	Dry matter yield $\text{g pot}^{-1}$	Micronutrient uptake, $\text{mg g}^{-1}$ dry wt.	
		Fe	Mn
Fe	3.8 a	1.80 a	0.25 a
$\text{Fe}_{50}$	4.4 b	2.13 b	0.22 a
$\text{Fe}_{100}$	4.0 ab	2.10 b	0.19 b
$\text{Fe}_{200}$	2.1 c	1.09 c	0.12 b
$\text{Mn}_0$	3.6 a	1.71 a	0.09 a
$\text{Mn}_{50}$	4.0 b	2.02 b	0.20 b
$\text{Mn}_{100}$	3.5 ab	1.85 ab	0.22 b
$\text{Mn}_{200}$	3.3 ac	1.57 a	0.26 c

\* Values not followed by the same letter in a column are significantly different.

Fe uptake in the plants increased significantly with applied Fe upto  $\text{Fe}_{100}$  and there after at  $\text{Fe}_{200}$  level the Fe uptake decreased significantly. Mn uptake increased with the successive rates of Mn application, whereas Fe application at  $\text{Fe}_{100}$  and above reduced the Mn uptake. There was a twofold decrease in Mn uptake with the application of  $\text{Fe}_{200}$ . However Mn application has an insignificant effect on the Fe uptake in plants (Table 1). Sommers and Shiev (1942) held that a higher concentration of soluble Fe in the tissue is invariably associated with a low concentration of soluble Mn, and vice versa. Alam (1985) also reported decreased Mn content in plants when Fe was applied beyond  $30 \text{ mg kg}^{-1}$  soil. This data suggested that a high level of Fe reduced the Mn content in the plants. Thus it appears that the main effect of Fe on Mn utilization by berseem is to reduce the rate of entry into the plant and induce Mn deficiency in plants. The effect of Fe application on Mn was much more pronounced than that of Mn of Fe.

#### *Soil pH and available Fe and Mn*

After the crop harvest, it was found that the original pH of the soil has decreased from 8.9 to 8.6. The ferrous sulphate application further decreased it to 8.2 and manganese sulphate application has negligible effect on soil pH (Table 2). The decrease in soil pH with  $\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$  application may be due to the reclamation effect of this fertilizer (Milap Chand et al. 1977).

The native soil Fe tended to increase with cropping (from 3.9 to  $4.2 \text{ mg kg}^{-1}$ ), whereas soil Mn has no effect. The increase in extractable Fe could

**Table 2**  
*Soil pH and recovery of added iron and manganese by DTPA extractant after the crop harvest*

Treatment	Soil pH	% Recovery	Mean
<i>DTPA extractable Fe</i> ( $\text{mg kg}^{-1}$ soil)			
$\text{Fe}_0$	8.6	4.3	—
$\text{Fe}_{50}$	8.5	6.3	4.2
$\text{Fe}_{100}$	8.4	11.9	7.7
$\text{Fe}_{200}$	8.2	19.6	7.7
			6.5
<i>DTPA extractable Mn</i> ( $\text{mg kg}^{-1}$ soil)			
$\text{Mn}_0$	8.6	1.7	—
$\text{Mn}_{50}$	8.6	8.5	13.6
$\text{Mn}_{100}$	8.5	12.7	11.0
$\text{Mn}_{200}$	8.5	27.0	12.0
			12.4



be attributed to the activity of micro-organisms and plant roots producing chelating agents which tend to hold more in solution (Pollett and Lindsay 1971).

The Fe and Mn fertilizers increased marginally their respective available content in soil (Table 2). This indicates that the residual value of these fertilizers in alkaline soils is very low. It also revealed that the recovery of added Fe after the crop harvest by DTPA soil test was only 6.5% (Table 2). The results agree with the reported findings of Follett and Lindsay (1971), that most of the soil applied inorganic Fe fertilizers ( $\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$ ) are rapidly fixed on soils and become highly unavailable. Thus the recovery of added Fe was only 20% in neutral soils after one week of its application.

Manganese fixation was less as compared to Fe as 12.4% of the added Mn was still extractable after 60 days of its application. Cook and Davis (1957) reported that Mn fertilizers rapidly oxide and precipitated as insoluble manganic oxides under neutral and alkaline conditions. The results agree with the findings of Pollett and Lindsay (1971), who reported that only 14% Mn is extractable by DTPA after 14 weeks of its addition to neutral soils. This study suggested that the higher rates of Fe application has an antagonistic effect on Mn availability in plants, whereas Mn has an insignificant effect on Fe availability for berseem crops.

## References

- Alam, S. M. (1985): Effect of iron and manganese on the growth of rice and on the contents of these elements in rice plants. *Agronomie*, **5**, 6, 487-90.
- Follett, R. N., Lindsay, W. L. (1971): Changes in DTPA-extractable zinc, iron, manganese, and copper in soils following fertilization. *Soil Sci. Am. Proc.*, **35**, 600-602.
- Gupta, U. C. (1972): Effect of manganese and lime on zinc and on the concentration of Mn, Mo, B, Cu and Fe in the boot stage tissue of barley. *Soil Sci.*, **114**, 131-36.
- Lindsay, W. L., Norvell, W. A. (1978): Development of DTPA soil test for zinc, iron, manganese and copper. *Soil Sci. Soc. Am. J.*, **42**, 421-28.
- Milap Chand-Abrol, I. P., Bhumbra, D. R. (1977): A comparison of the effect of eight amendments on soil properties and crop growth in a highly sodic soil. *Indian J. Agric. Sci.*, **47**, 348-54.
- Sideris, C. P., Young, H. Y. (1949): Growth and chemical composition of *Ananas comosus* in solution culture with different iron-manganese ratios. *Plant. Physiol.*, **24**, 416-40.
- Sommers, I. L., Shieve, J. W. (1942): The iron-manganese relation in plant metabolism. *Plant. Physiol.*, **17**, 582-602.
- Swarup, A., Mishra, M. (1972): Effect of ferrous sulphate on the availability of manganese. *J. Indian Co. Soil Sci.*, **20**, 417-19.
- Vorm, P. D., Van der, J., Diest Van A. (1979): Aspects of the Fe and Mn nutrition of rice plants. I. Iron and manganese uptake by rice plants, grown under aerobic and anaerobic conditions. *Pl. Soil.*, **51**, 233-46.
- Takkar, P. N., Nayyar, V. K. (1981): Preliminary field observations of manganese deficiency in wheat and berseem. *Fert. News*, **26**, 22-23 and 33.

## *Plant genetics and breeding*

### ABILITY OF VARIOUS MUTAGENS TO INDUCE CHROMOSOMAL ABERRATIONS IN PEAS (*PISUM SATIVUM* L.)

VO HUNG

DEPARTMENT OF PLANT GENETICS AND BREEDING, UNIVERSITY  
OF HORTICULTURE AND FOOD INDUSTRY, BUDAPEST, HUNGARY

(Received 1 June 1987; accepted 14 September 1987)

The aim was to study the cytological effect of various mutagens on different pea varieties, with special regard to the induction of chromosomal aberrations and the degree of aberration in various generations.

In treatments with 0.1% ethylenimine during mitosis metaphase, chromosome and chromatide fractures were observed.

In treatments with 0.5% ethyl methane sulphonate a low percentage of chromosomal aberrations was found in mitosis. No irregularities in division were observed in studies on meiosis.

In pea varieties treated with X-rays (5 kr) anomalies were found in cell division. The number of cells dividing abnormally rose parallel with the dosage.

Treatment with gamma rays in the  $M_1$  generation led to a high percentage of chromosomal aberrations in both mitosis and meiosis. Anomalies could still be observed in the  $N_2$  generation, but the frequency of chromosomal aberrations was substantially reduced compared to the  $M_1$  generation, while the  $M_3$  generation did not differ from the control.

In the radiation nursery there was a slight rise in the frequency of chromosomal aberrations in the  $M_1$  and  $M_2$  generations of the pea varieties tested after irradiation with  $^{60}\text{Co}$  compared with the control.

**Keywords:** chromosomal aberrations, ethylenimine, ethyl methane sulphonate, gamma-rays pea, X-rays

#### Introduction

It has long been known that certain mutagens may induce changes in chromosome structure, which means that chromosomal aberrations generally arise following mutagenic treatments.

Chromosomal aberrations are of great importance both in genetic research and in plant breeding practice, since chromosomal aberrations are easily identified and can be used to measure the efficiency of mutagenic agents.

Many literary data are available on the induction of chromosomal aberrations in peas. The aim of the present work is to determine the degree of chromosomal aberration arising in peas in mitosis and meiosis after treatment with various mutagens and the type of aberrations induced.



Certain authors found that the frequency of abnormal cell division was a function of radiation sensitivity (Monti et al. 1969, 1978, Sidorova et al. 1969, Gottschalk et al. 1970, Gottschalk 1981, Kaul 1978).

The frequency with which cytological aberrations arise after gamma radiation was analysed by Pershad et al. (1961), Akopyan (1967), Sidorova et al. (1967), Pyatenko (1970), Vo Hung (1972). It was observed that in peas the cytogenetic damage depends on the dosage of gamma rays. The larger the dosage, the greater the frequency of abnormal cell division.

A number of authors treated peas with X-rays and gamma radiation and obtained polyploids (Khvostova et al. 1959, Kurnik 1969).

After treatment with neutrons mitotic disturbances and aberrations arise in peas (Akoyan 1967, Tamássy et al. 1967).

The spectrum of chemical treatments becomes wider every year. Narsinghani et al. (1971) observed that EMS and MMS treatments inhibited the development of pea seedlings and induced chromosomal aberrations; this effect can be reduced by treatment with gibberellic acid.

Dudits (1967) treated pea seeds with EMS and observed that the frequency of aberrations changed as a function of the EMS concentration.

Zoz et al. (1965) found that the frequency of chromosomal aberrations was greater after treatment with NMK than after treatment with NEK.

The effect of a combined treatment with EMS and gamma rays was studied in peas by Mekhandiev et al. (1970), who reached the conclusion that the frequency of chromosomal aberrations decreased as a result of the combined physical and chemical treatments. The ratio of various types of aberrations depended on the treatment combination.

In the light of the above, the aim was to investigate the effect of the mutagens mentioned in the literature on cytological aspects, with special regard to the possible induction of chromosomal aberrations and to the extent of abnormality in various generations.

### Materials and methods

The experiments were set up at the Department of Plant Heredity and Breeding of the University of Horticulture.

The varieties "Gloria di Quimper", "Express", "Debreceni világoszöld" (round seeded peas), "Viridis", "Budai gyöngy and Erika" (marrowfat peas) were used in the investigations.

#### *Mutagen treatments*

(1) Physical treatment: dry seeds were treated with 5, 10, 15 and 20 kr doses of X-rays (1) and 5, 10, 15 and 20 kr doses of gamma radiation (2). The treatments were carried out in the Laboratory of the Isotope Institute of the Hungarian Academy of Sciences and the Laboratory of the National "Frédéric Joliot-Curie" Research Institute for Radiation Biology and Radiation Hygiene.



(2) Chemical treatment: dry seeds were treated for 12 hours with 0.05, 0.1 and 0.15% solutions of ethylenimine (EI) and 0.1, 0.3 and 0.5% solutions of ethyl methane sulphonate (EMS).  
(3) Sowing of pea varieties in the radiation nursery at Szigetcsép: Varieties: "Gloria di Quimper", "Express", "Debreceni világoszöld" (round seeded peas), "Viridis and Erika" (marrowfat peas). Source:  $^{60}\text{Co}$  with an activity of 150 Curie. Distance from source to plants: 5 m.

#### *Analytical methods*

The cytological analyses were carried out in mitosis and meiosis using the well-known rapid staining chromosome counting technique.

For the mitosis analyses root tips obtained from seeds germinated in Petri dishes were used. Approx. 25–30 root tips were studied per treatment and per variety. The material was fixed for 48 hours in Carnoy solution, then placed in a 75% alcohol solution. After a further 48 hours it was stained with carmine acetic acid, homogenated and in warmed 45% acetic acid.

For the meiosis analyses the material was picked prior to flowering, when the buds appeared, and examined using the technique described above. Buds were collected from approximately 20–25 plants per variety and per treatment.

### Results

Chromosome aberrations also occurred in the controls of the individual varieties, but to a negligible extent (0.51–0.87%). The chromosome number was  $2n = 14$  and  $n = 7$ .

The following changes were observed due to the effect of the different mutagens:

*Ethylenimine* (EI): in the 0.05% treatment the cell division in both mitosis and meiosis was close to normal. Anomalous cell division was found at 0.1%. Chromosome and chromatide fractures were observed in mitosis metaphase. Cell division in meiosis was normal.

*Ethyl methane sulphonate* (EMS): in the 0.1 and 0.3% treatments no significant cytological changes could be demonstrated in mitosis. In the 0.5% treatment, a low percentage of chromosome aberrations arose during mitosis in the varieties "Gloria di Quimper" and "Viridis". No abnormalities in cell division were found in the meiosis studies.

*X-rays*: abnormalities in cell division were noted even in pea varieties treated with 5 kr. As the dosage was increased, the number of cells with abnormal division also rose. After 10 and 15 kr doses of X-irradiation chromosomal aberrations were frequent (Table 1).

*Gamma rays*: a sensitive response came from the varieties "Gloria di Quimper" and "Erika", where cell division abnormalities were observed for all doses. Chromosome fractures were found in mitosis metaphase and chromosome bridges and chromatide fractures in anaphase.

Severely fragmented chromosomes were observed in mitosis metaphase after irradiation with 15 kr. In ana- and telophase some of the damaged chromosomes remained in the equatorial plane.

(1) and (2): 80 r/min intensity



**Table 1**

*Chromosomal aberrations induced by mutagenic treatments in mitosis*

Treatment	No. of dividing cells	Fragmentation (%)	Bridges (%)	Total chromoso- mal aberrations (%)	No. of dividing cells
(1)	(2)	(3)	(4)	(5)	(6)
Variety: "Gloria di Quimper"					
Control	285	$0.70 \pm 0.3$	0.00	0.70	228
X 5kr	198	$4.04 \pm 1.2^*$	0.00	4.04	270
X 10kr	381	$10.23 \pm 2.0^{***}$	$1.04 \pm 0.8$	11.27	122
X 15kr	155	$27.09 \pm 2.4^{***}$	$1.93 \pm 1.0$	29.02	95
Gamma 5kr	324	$6.48 \pm 2.5^*$	$1.85 \pm 1.0$	8.33	441
Gamma 10kr	372	$19.08 \pm 2.1^{***}$	$2.41 \pm 0.9^*$	21.49	236
Gamma 15kr	214	$35.04 \pm 1.9^{***}$	$3.73 \pm 1.0^{**}$	38.77	255
Gamma 20kr	191	$50.78 \pm 2.6^{***}$	$2.61 \pm 0.6^{**}$	53.39	—
EI 0.05%	96	$1.04 \pm 0.6$	0.00	1.04	144
EI 0.1%	72	$11.11 \pm 2.2^{***}$	$1.38 \pm 0.9$	12.49	105
EMS 0.1%	104	$0.96 \pm 0.5$	0.00	0.96	75
EMS 0.3%	81	$1.23 \pm 0.6$	0.00	1.23	122
EMS 0.5%	66	$10.60 \pm 2.3^{**}$	$1.51 \pm 0.8$	12.11	48
Variety "Erika"					
		M <sub>1</sub>			
Control	196	$0.51 \pm 0.4$	0.00	0.51	174
X 5kr	250	$8.00 \pm 1.4^{***}$	$0.40 \pm 0.3$	8.40	108
X 10kr	102	$13.72 \pm 1.2^{***}$	$2.94 \pm 1.0^{**}$	16.66	224
X 15kr	84	$28.57 \pm 2.0^{***}$	$2.38 \pm 1.1^*$	30.95	145
Gamma 5kr	235	$4.68 \pm 1.7^*$	0.00	4.68	312
Gamma 10kr	114	$16.66 \pm 1.8^{***}$	$3.50 \pm 1.4^*$	20.16	226
Gamma 15kr	182	$18.68 \pm 2.6^{***}$	$5.49 \pm 1.5^{***}$	24.17	234
EI 0.05%	66	$1.51 \pm 0.7$	0.00	1.51	112
EI 0.1%	92	$13.04 \pm 1.0^{***}$	$2.17 \pm 1.1^*$	15.21	81
EMS 0.15%	156	$0.64 \pm 0.4$	0.00	0.64	48
EMS 0.3%	55	$1.81 \pm 0.6$	0.00	1.81	102
EMS 0.5%	87	$6.89 \pm 1.6^{**}$	$1.14 \pm 0.7$	8.03	72

\*: Difference significant at the P = 5% level

\*\*: Difference significant at the P = 1% level

\*\*\*: Difference significant at the P = 0.1% level

*(n = 10 preparations) 1985–1986–1987*

Fragmentation (%)	Bridges (%)	Total chromosomal aberrations (%)	No. of dividing cells	Fragmentation (%)	Bridges %	Total chromosomal aberrations (%)
(7)	(8)	(9)	(10)	(11)	(12)	(13)
0.87±0.5	0.00	0.87	148	0.00	0.00	0.00
1.11±1.0	0.00	1.11	77	1.29±0.6	0.00	1.29
2.45±1.1	1.63±0.8	4.08	204	0.98±0.5	0.00	0.98
4.21±1.2*	2.10±1.0	6.31	60	1.66±0.7	0.00	1.66
1.17±0.8	0.29±1.2	1.46	122	0.81±0.4	0.81±0.4	1.62
3.38±1.0*	1.69±0.9	5.07	85	1.17±0.5	0.00	1.17
5.49±1.8*	1.96±0.8	7.45	151	1.32±0.7	0.00	1.32
—	—	—	—	—	—	—
0.69±0.4	0.00	0.69	65	1.53±0.9	0.00	1.53
1.90±1.0	0.00	1.90	116	1.72±0.8	0.00	1.72
1.33±0.6	0.00	1.33	98	1.02±0.6	0.00	1.02
1.63±0.8	8.81±0.5	2.44	140	0.71±0.4	0.71±0.4	1.42
2.08±1.1	0.00	2.08	86	1.16±0.8	0.00	1.16
<b>M<sub>2</sub></b>				<b>M<sub>3</sub></b>		
0.00	0.00	0.00	188	0.53±0.3	0.00	0.53
0.00	0.00	0.00	116	0.86±0.5	0.00	0.86
1.33±1.0	0.00	1.33	81	1.23±0.8	0.00	1.23
2.75±1.2*	0.68±0.4	3.43	66	1.51±0.9	0.00	1.51
0.00	0.00	0.00	204	0.98±0.5	0.49±0.3	1.47
2.22±1.2	0.44±0.3	2.66	115	0.86±0.4	0.00	0.86
3.71±1.6*	1.23±0.8	4.94	70	1.42±0.5	0.00	1.42
0.88±0.4	0.00	0.88	95	1.05±0.4	0.00	1.05
1.23±0.8	0.00	1.23	52	1.92±0.8	0.00	0.92
2.08±1.1	0.00	2.08	78	1.28±0.5	1.28±0.5	2.56
1.96±1.0	0.00	1.96	134	1.49±0.7	0.00	1.49
2.77±1.8	1.39±0.9	4.16	97	1.03±0.5	0.00	1.03



In 10 and 15 *kr* treatments abnormal cell division was also experienced in meiosis. In the first metaphase of meiosis the separation of the bivalents was protracted, as the result of which chromosomal aberrations arose.

In the 20 *kr* treatment the cells were damaged to such an extent that the seedlings were unable to continue their development and died.

In the  $M_2$  generation the cytological examinations were repeated in mitosis and meiosis and the following conclusions were drawn:

Abnormal cell division was repeatedly observed as the result of gamma radiation in the varieties "Gloria di Quimper" and "Erika". The experimental results indicate that cell division proceeds as normal at gamma ray doses

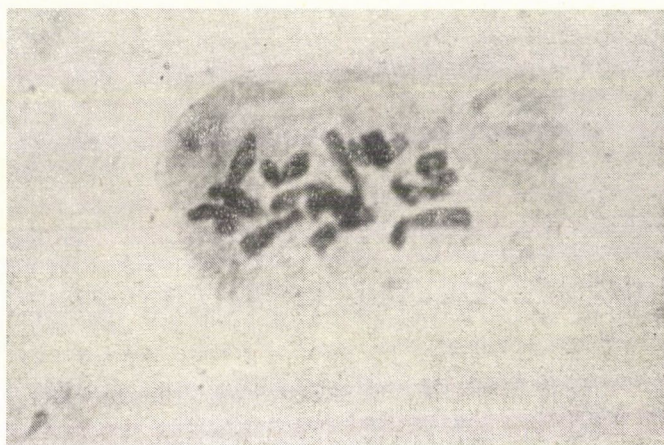


Fig. 1. Control, mitosis, metaphase,  $2n = 14$  (1800  $\times$ )

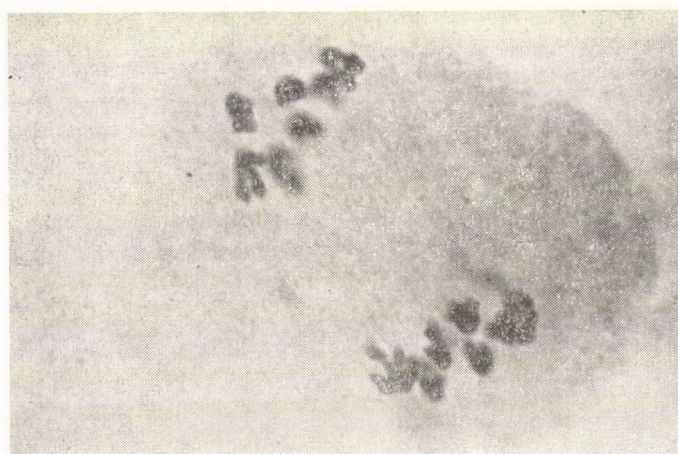


Fig. 2. Control, meiosis I, anaphase,  $n = 7$  (1800  $\times$ )



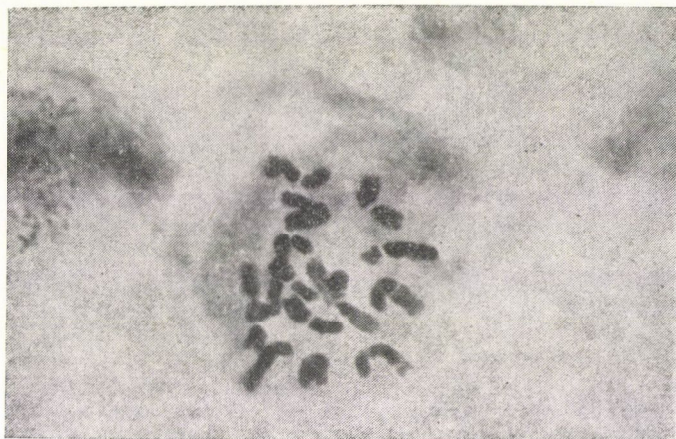


Fig. 3. Mitosis, metaphase, fragmented chromosomes (X-rays, 10 kr) (1800  $\times$ )



Fig. 4. Mitosis, anaphase, lagging chromosomes (gamma rays, 15 kr) (1800  $\times$ )

of 5 and 10 kr. A dose of 15 kr led to fragmented chromosomes in mitosis metaphase and to chromosome and chromatide bridges in anaphase.

In the first metaphase and anaphase of meiosis chromosome and chromatide bridges and rings (translocations) arose.

*Mutagenic treatments* in the  $M_1$ ,  $M_2$  and  $M_3$  generations caused chromosomal aberrations in the varieties "Gloria di Quimper" and "Erika". The percentage of aberrations was calculated in mitosis and can be seen in Table 1, from which it is clear that the higher the dosage, the larger the number of abnormal cells that appear.





Fig. 5. Meiosis I, anaphase, chromosome bridges (gamma rays, 15 kr) (1800 × )

When comparing the frequency of chromosomal aberrations in the three generations, for the variety "Gloria di Quimper" for instance, it is found that while the rate of aberrations as the results of a gamma ray dose of 15 kr was 38.77% in the  $M_1$  generation, this rate was 7.45% for  $M_2$  plants originating from the  $M_1$  treatment and only 1.32% for  $M_3$  plants, indicating that in the course of development the changed cell lines are selected out and the cells divided normally from then on.

Table 2

*Frequency of abnormal cell division in meiosis after  $Co^{60}$  irradiation (radiation nursery) (n = 10 preparations) 1985-1986*

Treatment	M <sub>1</sub>			M <sub>2</sub>		
	No. of dividing cells	Abnormal cell No.	division %	No. of dividing cells	Abnormal cell No.	division %
	Variety: "Gloria di Quimper"					
Control (field experiment)	112	1	0.89±0.5	82	0	0.00
Radiation nursery	78	3	3.84±1.1	94	3	3.19±1.0
	Variety: "Viridis"					
Control	68	0	0.00	45	0	0.00
Radiation nursery	55	2	3.63±1.0	No cell division		
	Variety: "Erika"					
Control	32	0	0.00	No cell division		
Radiation nursery	104	4	3.84±1.2	64	2	3.12±0.9

In the  $M_3$  generation there was no significant difference between the control and the treated material as regards the frequency of chromosomal aberrations.

It is also clear from Table 1 that fragmentation was more frequent than anaphase bridges. There was no significant difference between the two varieties in the frequency of aberrations.

Frequency of chromosomal aberrations induced by  $^{60}\text{Co}$  irradiation (in the radiation nursery) in meiosis in the  $M_1$  and  $M_2$  generations: cytological analyses were carried out in the meiosis of  $M_1$  and  $M_2$  generations of a number of pea varieties grown in the radiation nursery. The following results were obtained:

No significant difference was found between the  $M_1$  and  $M_2$  generations with respect to the frequency of chromosomal aberrations. For the pea varieties studied, however, there was a slight rise in the frequency of chromosomal aberrations in the  $M_1$  and  $M_2$  generations after irradiation with  $^{60}\text{Co}$  compared to the control (Table 2).

### Summary

The aim was to study the cytological effect of certain mutagens on various pea varieties, with special regard to the induction of chromosomal aberrations and the rate of abnormality in different generations.

Summarizing the results, the following conclusions can be drawn:

(1) After a 0.1% treatment with ethylenimine, abnormal cell division was found in mitosis. In mitosis metaphase chromosome and chromatide fractures were observed. In meiosis cell division was normal.

(2) In the case of treatment with 0.5% ethyl methane sulphonate, a low percentage of chromosomal aberrations arose in mitosis. No anomalies in cell division were found in studies on meiosis.

(3) In pea varieties treated with X-rays, even a dose of 5 kr led to abnormalities in cell division. As the dosage increased, there was a parallel rise in the number of cells undergoing abnormal division.

(4) In the case of gamma irradiation a high rate of chromosomal aberrations was recorded in both mitosis and meiosis in the  $M_1$  generation. The abnormalities were still demonstrable in the  $M_2$  generation, but the frequency of chromosomal aberrations was substantially reduced compared to the  $M_1$  generation, while the  $M_3$  generation did not differ from the control.

(5) In pea varieties treated with  $^{60}\text{Co}$  irradiation in the radiation nursery, the frequency of chromosomal aberrations was slightly higher in the  $M_1$  and  $M_2$  generations than in the control.



### References

- Dudits, D. (1967): Az indukált mutációs folyamatok vizsgálata borsó növényeken (Study of induced mutational processes in pea plants). Doctoral dissertation.
- Kurnik, E. (1969): Borsó, In: Magyar Növénynevelés (Peas. In: Hungarian Plant Breeding). Akad. Kiadó, Budapest, 341–358.
- Tamássy, I., Nagy-Porpáczy, B., Hoppán, G. (1967): Növekedés-szabályozó és mutagén anyagok hatásának citológiai vizsgálata hímsteril borsóformák előállítására (Cytological study on the effect of male sterile pea forms). *Növénytermelés*, **16**, 2, 131–140.
- Vo Hung (1972): Gamma besugárzás hatásának citológiai vizsgálata borsónál (Cytological studies on the effect of gamma irradiation in peas). *A Kertészeti Egyetem Közleményei*, **36**, 183–191.

## INHERITANCE OF FLAG-LEAF AREA IN TWO INTERVARIETAL CROSSES OF BREAD WHEAT (*TRITICUM AESTIVUM* L.)

J. S. BIJRAL, K. S. KANWAL, B. GUPTA, B. SINGH and  
T. R. SHARMA

DIVISION OF PLANT BREEDING AND GENETICS, REGIONAL  
AGRICULTURAL RESEARCH STATION  
R. S. PURA (JAMMU — INDIA)

(Received 20 July 1987; accepted 30 November 1987)

Parents with different generations ( $F_1$ ,  $F_2$ ,  $B_1$  and  $B_2$ ) of two intervarietal crosses of bread wheat viz., VL 421  $\times$  RSP 37 and HD 2307  $\times$  Kalyansona were used for the estimation of genetic components for flag-leaf area. A comparison of the generation means showed  $F_1$  to be either intermediate between the two parents or significantly superior to the better parent. Both additive and additive  $\times$  dominance components of genetic effects were important for this character and the observed epistasis was of complementary type.

**Keywords:** wheat, *Triticum aestivum* L., intervarietal crosses, flag-leaf area

### Introduction

Genetic manipulations for an optimum assembly of yield components have played a very vital role in grain improvement, but comparatively much less attention seems to have been paid to an important morpho-physiological trait like flag-leaf area which is reported to contribute about 42% of total grain yield in wheat (Mackey, 1982). A high degree of association between grain yield and flag-leaf area has also been reported by Yap and Harvey (1972); Monyo and Whittington (1973); Kraljevic-Balalic (1974); Barriaga and Fulntealba (1976); Briggs and Aytenfisu (1980) and Joshi et al. (1982). However, the genetic information on this trait is meagre. The present study was therefore undertaken to investigate the nature of gene action involved in the expression of flag-leaf area.

### Materials and methods

The material of the present investigation comprised two  $F_1$ s viz., VL 421  $\times$  RSP 37 and HD 2307  $\times$  Kalyansona, their parents, backcrosses and  $F_2$ s. The planting arrangement was a randomized block design with three replications. The distance between and within rows was 30 and 10 cm, respectively. The parents and  $F_1$ s were represented by single rows, whereas  $F_2$ s and backcrosses were represented by 10 rows/cross. Ten competitive plants each from parents,  $F_1$ s and backcrosses, and 40 plants from  $F_2$  generations were selected at random for



recording flag-leaf area. The flag-leaf area was estimated by the formula as suggested by Simpson (1968). The estimates of genetic parameters were calculated following Cavalli (1952) and Hayman (1958).

## Results and discussion

The analysis of variance (Table 1) indicated highly significant differences among different generations of both the crosses for flag-leaf area. A study of the means of different generations revealed that the  $F_1$  was intermediate between the two parents in VL 421  $\times$  RSP 37, whereas in HD 2307  $\times$  Kalyan-

**Table 1**  
*Mean performance of different generations and analysis of variance for flag-leaf area (cm<sup>2</sup>)*

Generations/Mean squares	Crosses	
	VL 421 $\times$ RSP 37	HD 2307 $\times$ K. Sona
P <sub>1</sub>	41.76	46.70
P <sub>2</sub>	57.69	46.09
F <sub>1</sub>	49.53	51.09
F <sub>2</sub>	44.00	47.85
B <sub>1</sub>	37.35	58.68
B <sub>2</sub>	56.24	39.88
Replication mean squares	2.40	2.49
Generation mean squares	119.20**	116.34**
Error mean squares	5.99	8.37
LSD (0.05)	3.60	4.27

\*\* Significant at 1% level of probability

**Table 2**  
*Estimates of three genetic parameters based on different generation means for flag-leaf area (cm<sup>2</sup>)*

Genetic parameters	Crosses	
	VL 421 $\times$ RSP 37	HD 2307 $\times$ K. Sona
m	49.45** + 1.75	39.12** + 1.59
(d)	— 5.64** + 1.76	10.79** + 1.60
(h)	— 5.24** + 2.60	13.27** + 1.85
$Z^2(3)$	230.13**	61.34**

\* Significant at 5% level of probability

\*\* Significant at 1% level of probability

Table 3

*Estimates of the additive, dominance and interaction parameters based on different generation means for flag-leaf area (cm<sup>2</sup>)*

Genetic parameters	Crosses	
	VL 421 × RSP 37	HD 2307 × K. Sona
m	44.00** + 1.01	47.84** + 2.32
(d)	-18.89** + 2.52	18.79** + 1.93
(h)	10.97 + 6.63	10.42 + 10.13
(i)	11.16 + 6.45	5.73 + 10.05
(j)	-10.92** + 2.59	18.48** + 2.24
(l)	0.16 + 11.27	7.88 + 12.35

\*\* Significant at 1% level of probability

sona, the  $F_1$  performance was significantly higher than that of the better parent, indicating dominance for this character. It is, however, interesting to note that the  $F_2$  means were almost comparable to the  $F_1$  means. This indicated that there was not much inbreeding depression and the expression obtained in the  $F_1$  is likely to be maintained.

The analysis of generation means on an additive-dominance model was done on six generations. Since the  $\chi^2$  values were found to be highly significant for the trait under study in both the crosses (Table 2), the data were extended to a digenic epistatic model.

Estimates of additive, dominance and interaction parameters are presented in Table 3. The data indicated that additive, as well as additive × dominance (j) components of epistasis, were highly significant in both the crosses and the observed epistasis was of complementary type. Almost similar results have also been reported by Joshi and Sharma (1984).

The significance and preponderance of additive and epistatic (additive × dominance) types of gene effects in both the crosses suggests that simple breeding procedures coupled with recurrent selection involving cyclic intermating of the selects would be the most desirable selection procedure for the simultaneous exploitation of fixable and non-fixable gene action (Jensen, 1970; Reddne and Jensen, 1974; and Wright, 1980). Because intermating in early generations is likely to break the undesirable linkage, it may subsequently result in the establishment of rare useful recombinant.

## References

- Barriga, b. P., Fulntealba, A. J. (1976): Relations between yield, yield components and morphological characters above the flag-leaf node in spring wheat. *Agron. Sur.*, **4**, 650-670.  
 Briggs, K. G., Aytenfisu, A. (1980): Relationship between morphological characters above the flag-leaf node and grain yield in spring wheat. *Crop Sci.*, **20**, 350-354.



- Cavalli, L. L. (1952): *An analysis of linkage in quantitative inheritance*. (In: Quantitative Inheritance, ed. E. C. R. Rieve and C. H. Waddington), 135-144. HM 50, London.
- Hayman, B. I. (1958): The separation of epistatic from additive and dominance variation in generation means. *Heredity*, **12**, 371-390.
- Jensen, N. F. (1970): A diallel selective mating system for cereal breeding. *Crop Sci.*, **10**, 629-635.
- Joshi, A. K., Sharma, G. S., Dhari, R. (1982): Variability and associations of flag-leaf area and other traits in wheat. *Indian J. Agric. Sci.*, **52**, 351-55.
- Joshi, A. K., Sharma, G. S. (1984): Genetics of flag-leaf area in wheat-triallel analysis. *Ind. J. Genet.* **44**, 399-405.
- Kraljevic-Balalic, M. (1974): Inheritance of leaf area in vulgare wheat. *Wheat Inf. Service*, **38**, 5-7.
- Mackey, J. (1982): *Shoot: root interrelation in cereals and its impact on the ideotype concept*. Proc. International Symp. on new genetical approaches to crop improve. Held at Karachi, Pakistan (in press).
- Monyo, J. R., Whittington, W. J. (1973): Genotypic differences in flag-leaf area and their contribution to grain yield in wheat. *Euphytica* **22**, 600-606.
- Reddne, R. J., Jensen, N. F. (1974): Mass selection and mating systems in cereals. *Crop Sci.*, **14**, 345-350.
- Simpson, G. M. (1968): Association between grain yield per plant and photo-synthetic area above the flag-leaf node in wheat. *Can. J. Plant Sci.*, **48**, 253-260.
- Wright, A. J. (1980): The expected efficiency of half sib, test cross and SI progeny testing methods in single population improvement. *Heredity*, **45**, 361-376.
- Yap, T. C., Harvey, B. L. (1972): Inheritance of yield components and morpho-physiological traits in barley. *Crop Sci.*, **12**, 283-86.

## *Plant protection*

# COMPARATIVE STUDY OF THE PHYTOTOXICITY OF ACETANILIDE HERBICIDES ON MAIZE (*ZEA MAYS* L.) AS AFFECTED BY TEMPERATURE AND ANTIDOTES

Z. BERZSENYI and B. GYÖRFFY

AGRICULTURAL RESEARCH INSTITUTE OF THE HUNGARIAN ACADEMY OF SCIENCES,  
MARTONVÁSÁR, HUNGARY

(Received: 26 October 1987)

The phytotoxicity of propachlor, alachlor, metolachlor and acetochlor as well as the effect of acetochlor-antidote combinations were studied in growth (PGV 36 type) and gradient phytotron chambers on maize (*Zea mays* L.). A continuous linear temperature gradient was maintained in the gradient chambers (between 13-22 °C). Herbicide rates were 1, 2, 4, 8 and 16 l a.i./ha. the antidotes AD-67 (N-dichloroacetyl-l-oxa-4-azasprio-4,5-decane) and DKA-24 (N, N<sup>2</sup>-diallyl-N<sup>2</sup>-dichloroacetyl-glycinamide) were used in a 10 : 1 (w/w) ratio.

In the growth (homogeneous) chamber experiments, the phytotoxic effect of the acetanilides was characterized by the fresh weight and plant height data expressed as a percentage of the control. The phytotoxic effect was assessed by scoring with a 1-5 scale. In the gradient chambers the dependence of treatments on temperature was characterized using a linear function fitted to the data pairs. An increase of the regression coefficient (b) indicated greater phytotoxic injury, whereas in the acetochlor-antidote combinations a stronger antidote effect was noted.

It was found that the phytotoxicity of the acetanilide herbicides tested increased in the following order:

propachlor → alachlor → metolachlor → acetochlor

Their weed control effect exhibited an opposite tendency. The phytotoxic effect of propachlor did not increase significantly up to the 16 l/ha dose. A definite dosage effect could be observed in the phytotoxicity of herbicides alachlor, metolachlor and acetochlor.

The experimental data did not indicate that an increase in temperature from 13-22 °C had any significant effect on phytotoxic injury to maize plants. Further experiments are required for the exact investigation of this problem. Our experimental data verify that antidotes should be added to the less selective acetanilide herbicides (e.g. acetochlor, metolachlor). Antidotes AD-67 and DKA-24 had reliable protective effects; however, in the case of overdosage of acetochlor and metolachlor (4 l a.i./ha), the antidote effect decreased.

**Keywords:** acetanilide herbicides, maize, temperature, antidotes, *Zea mays* L.

## Introduction

The demonstration of the weed control effect of acetanilide herbicides in 1952 yielded a similar development in the history of weed control as did the discovery of 2,4-D in 1944 (Hamm, 1974). Acetanilides control mono-



cotyledonous as well as some dicotyledonous weeds selectively in both mono- and dicotyledonous crops. Their weed control effect is based on the inhibition of protein and nucleic acid synthesis (Dean et al. 1980, Duke et al., 1975, Rao-Duke, 1976). They inhibit seedling development, especially root growth, but do not reduce seed germination. They are primarily translocated through the apoplast. In grasses, absorption takes place mainly through the coleoptile, whereas in broad-leaved weeds, penetration through the roots was discovered more important (Chandler et al. 1974). Both sensitive (e.g. wheat) and tolerant (e.g. maize) plant species are able to break down acetanilides; however faster detoxification was observed in tolerant plants (Jablonkai and Dutka 1985). The decomposition of acetanilides is in 90% a microbiological one, thus their half-life is relatively short.

This class of herbicides contains those with different weed control effects and crop selectivities. Among them alachlor, metolachlor and propachlor have been used for more than two decades in the weed control of maize in various countries. In Hungary acetochlor is widely used for pre-emergent weed control in maize. The first weed control experiments with acetochlor were conducted at Martonvásár in the late 60s. The experiments of Györfy (1977) proved that the weed control effect of acetochlor regarding both grasses and broad-leaved weeds was better than that of propachlor or alachlor. Its superiority was especially significant in years with low precipitation. Its great advantage is that it is effective against the atrazine-resistant biotype of *Amaranthus retroflexus* L.

The selectivity of acetanilide herbicides is influenced by herbicide phytotoxicity, method of application, herbicide-soil interactions and climatic factors. Several factors (formulation, morphological differences, absorption, translocation) affect herbicide concentration in plants. The most important, however, are herbicide metabolism and detoxification, as a result of which herbicide concentration decreases below the level of injury.

Several authors have demonstrated that acetanilide herbicides differ significantly in their phytotoxic effect. Among the five acetanilide herbicides studied by Leavitt and Penner (1978), alachlor was the least phytotoxic and did not injure the maize plant, up to a rate of 13.4 kg/ha in glasshouse experiments. The phytotoxicity investigations of Györfy (1977) proved that maize tolerated propachlor the best, whereas its tolerance to acetochlor and metolachlor was similar.

The phytotoxic injury of acetanilides is significantly influenced by the quantity of precipitation. The injury to maize plants increases if the herbicide is washed into a deeper, critical soil layer before maize emergence. Among the environmental factors, the effect of temperature on the phytotoxicity of acetanilides is less known. Experiments with alachlor on oat and bean plants revealed an increase in injury ranging between 10–17 °C and 20–25 °C



respectively. However, higher temperatures did not increase damage (Mulder and Nalevaja 1978; Penner and Graves 1972).

Selective chemical weed control reached a new phase with the development of chemical antidotes (safener, protectant) which selectively protect crops from herbicide injury without decreasing the weed control effect of the herbicide (Pallos and Casida 1978, Gimesi 1981). Chang et al. (1973) revealed that the antidote R-25788 (N,N-diallyl-2,2-dichloroacetamide) was more effective in protecting maize from thiocarbamate herbicide injury than from alachlor herbicide injury. In the experiments of Leavitt and Penner (1978), as well as Winkle et al. (1980) among the examined six potential antidotes, the protective effect of the antidote R-25788 acted best against alachlor, metolachlor and other acetanilide herbicide injuries to maize.

Phytotron and field experiments with various antidotes have now been conducted at Martonvásár for several years in order to increase acetochlor selectivity. Partly as a result of these experiments, acetochlor is now produced nearly exclusively in the antidoted form. The purposes of the study were: (1) to compare the phytotoxic effects of propachlor, alachlor, metolachlor and acetochlor on maize under exact phytotron conditions, (2) to examine the effects of different temperatures on herbicide selectivity and (3) to determine the protective effect of antidotes against acetochlor injury.

### Materials and methods

The experiments were set up under conditions which could be programmed and reproduced in so-called homogeneous (PGV 36 type) and gradient growth chambers in 1986. In the PGV chambers the day temperature was set at 18 °C, and the night temperature at 13 °C, whereas the relative humidity was 80%. The length of the photoperiod was 16 hours, and the intensity of illumination 30 klux. In the gradient chamber the effect of temperature on herbicide selectivity and the protective effect of antidotes was studied under a continuous linear temperature gradient (13.4; 14.2; 14.9; 15.4; 16.5; 17.5; 18.5; 19.7; 20.8; 21.8 °C). The length of the photoperiod was 16 hours, the intensity 30 klux, and the relative humidity 80%.

The experiments were arranged as follows, using a standard method: A mixture of meadow loam and sandy soil (1 : 1 ratio) was placed in the pots. In the homogenous chambers 18 and in the gradient chambers 12 seeds were planted per pot, 2 cms deep. After planting, the herbicides were applied using a special laboratory sprayer. The quantity of water required for the optimal development of plants was applied in the gradient chambers on the basis of weight measurements, whereas in the PGV chambers the same quantity of water was given per pot. The length of the experiments was 21 days in the PGV chambers and 16 days in the gradient chambers.

Each experiment was set up at least twice and there were 4–6 replications within each experiment. The treatments were randomized in the growth chambers and the pots were repositioned every 3–4 days. The measurements, and observations were made per plant, and the data expressed as an average of the plant number per pot were the units of observation.

The phytotoxic injury caused by the herbicides was assessed by scoring on the 14th and 21st day using a 1–5 scale (1 = no injury, 5 = lethal injury). At the end of the experimental period plant height, shoot — and in certain experiments — root weight were measured. Plant height meant the distance from the soil surface to the tip of the highest developed leaf. Herbicides propachlor, alachlor, metolachlor and acetochlor were tested in the experiments at rates of 1, 2, 4, 8 and 16 ai./ha. In the acetochlor — antidote combinations the antidotes



marked AD-67 and DKA-24 were used at a 10 : 1 (w/w) ratio. The chemical composition of the antidotes is as follows: AD-67 = N-dichloroacetyl-1-oxa-4-azaspiro-4,5-decane, DKA-24 = N, N<sup>2</sup>-diallyl-N<sup>2</sup>-dichloroacetyl-glycinamide (Görög et al. 1982, Nagy and Balogh, 1985).

The experiments — depending on the purpose of the tests — were set up with one or more maize hybrids. In identical types of experiments the same hybrids were planted in every case. The following hybrids were included in the experiments: "Mirna" (Pi 3925 SC), "Dea" (Pi 3839 SC), "NK PX" 9283 SC and "Pi3709 MSC".

The biometric evaluation of the experiments arranged in the PGV phytotron chambers was done using analysis of variance. In the case of the experiments conducted in the gradient chambers, the dependence of treatments on temperature was characterized by a linear function fitted to the data pairs. The regression coefficients of the linear functions were compared to determine the differences in herbicide selectivity and antidote effect.

## Results

### *Comparison of the phytotoxic effect of acetanilide herbicides under identical temperatures and at various rates*

Figure 1 shows the fresh weight and plant height data of three-week maize plants as affected by the rate of the tested acetanilide herbicides expressed as a percentage of the control. Table 1 summarizes the phytotoxic injury scores assessed on two occasions.

It was found that the phytotoxic effect of propachlor was very slight and did not reach the significant level. At the same time the phytotoxic effect of propachlor did not increase within the dosage limits examined. On the other hand, with the herbicides alachlor, metolachlor and acetochlor a definite dosage effect relationship could be observed in the extent of phytotoxic injury. At a rate of 1 l/ha the phytotoxic effect of alachlor and metolachlor is small (the fresh weight expressed as a percentage of the control was

Table 1  
*Degree of phytotoxic injury to maize plants in the 2nd and 3rd week following herbicide treatment*

Dose, l/ha	Propachlor		Alachlor		Metolachlor		Acetochlor	
	Number of weeks after treatment							
	2	3	2	3	2	3	2	3
Phytotoxic injury								
0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0
1	1.2	1.1	1.5*	1.2*	1.6*	1.6*	3.9*	3.1*
2	1.1	1.2	2.7*	1.9*	3.5*	2.9*	4.7	3.9*
4	1.1	1.0	3.3	2.4*	4.4	4.1	4.9	4.6
8	1.1	1.1	3.8	3.1	4.7	4.6	5.0	4.8
L.S.D (P=0.05%)	NS	NS	0.4	0.3	0.4	0.3	0.4	0.3

+ Score: 1 = no injury; 5 = every plant has died;

\* significantly different at P<sub>5</sub>%

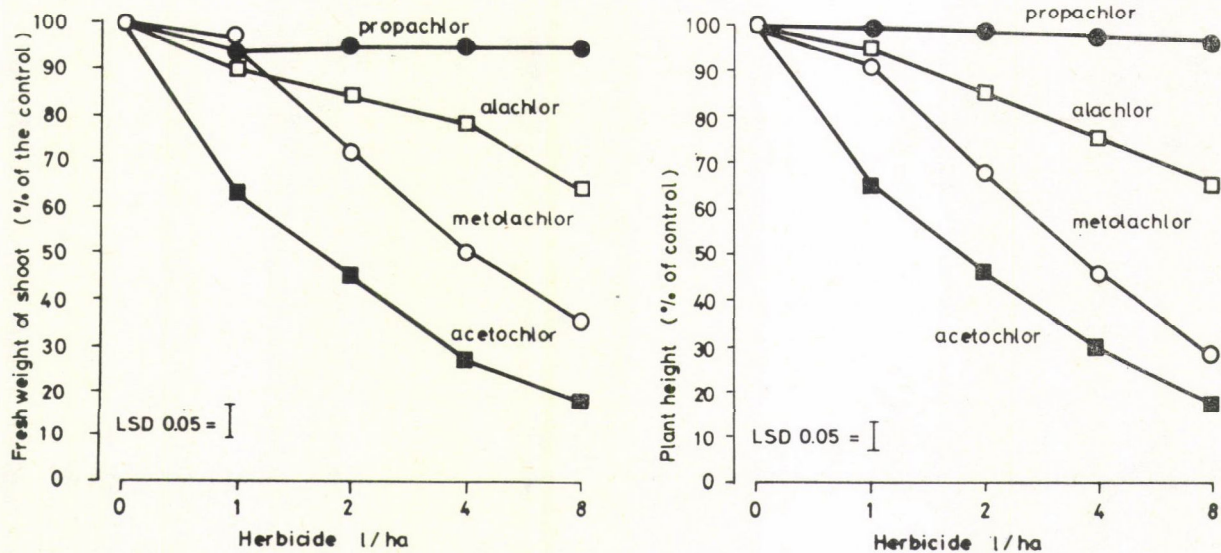


Fig. 1. Comparison of the phytotoxic effect of acetanilide herbicides as affected by the rate of application



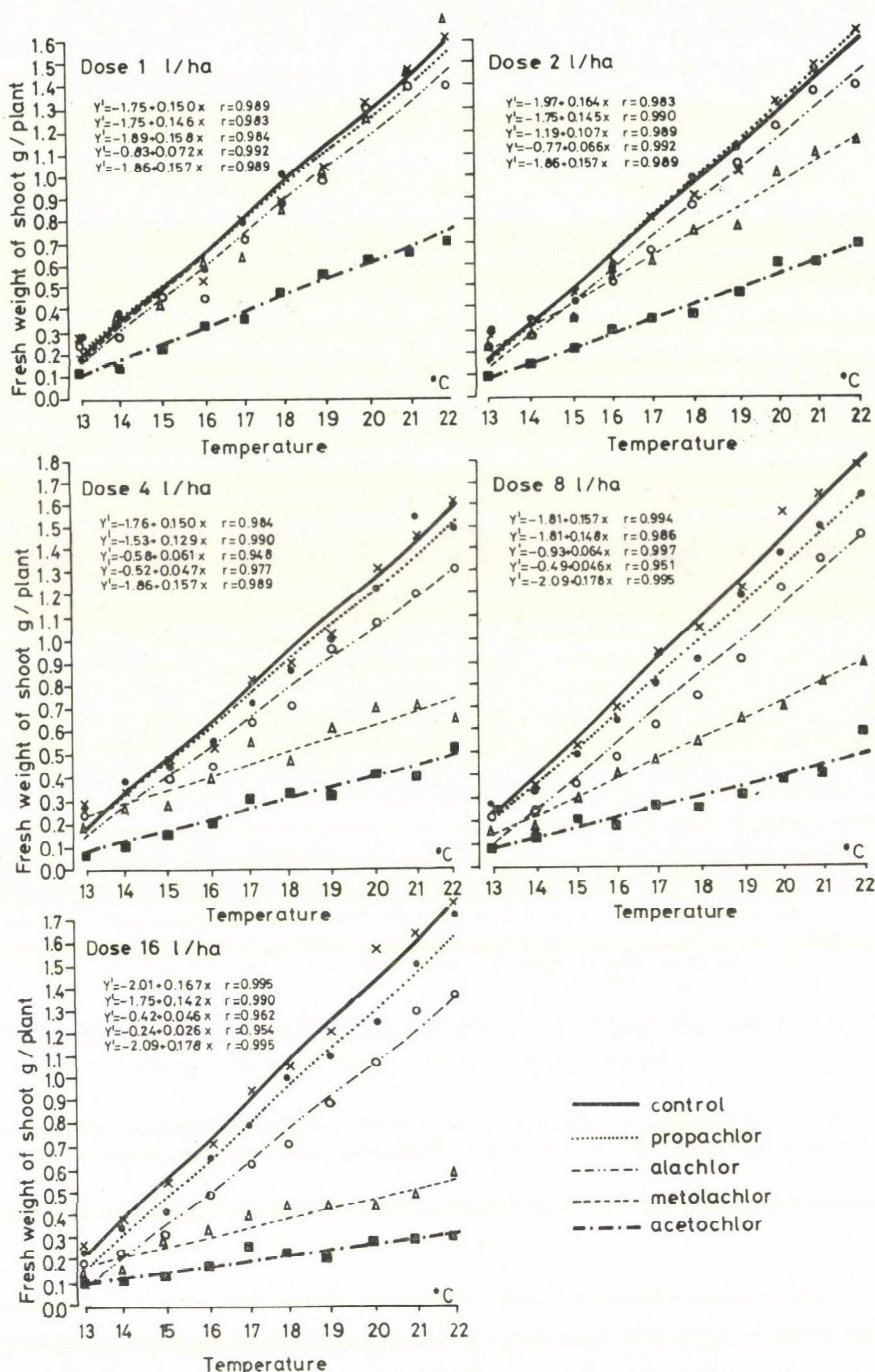


Fig. 2. Effect of different rates of the examined 4 chloroacetanilide herbicides on the fresh shoot weight of the maize plant in a gradient phytotron experiment

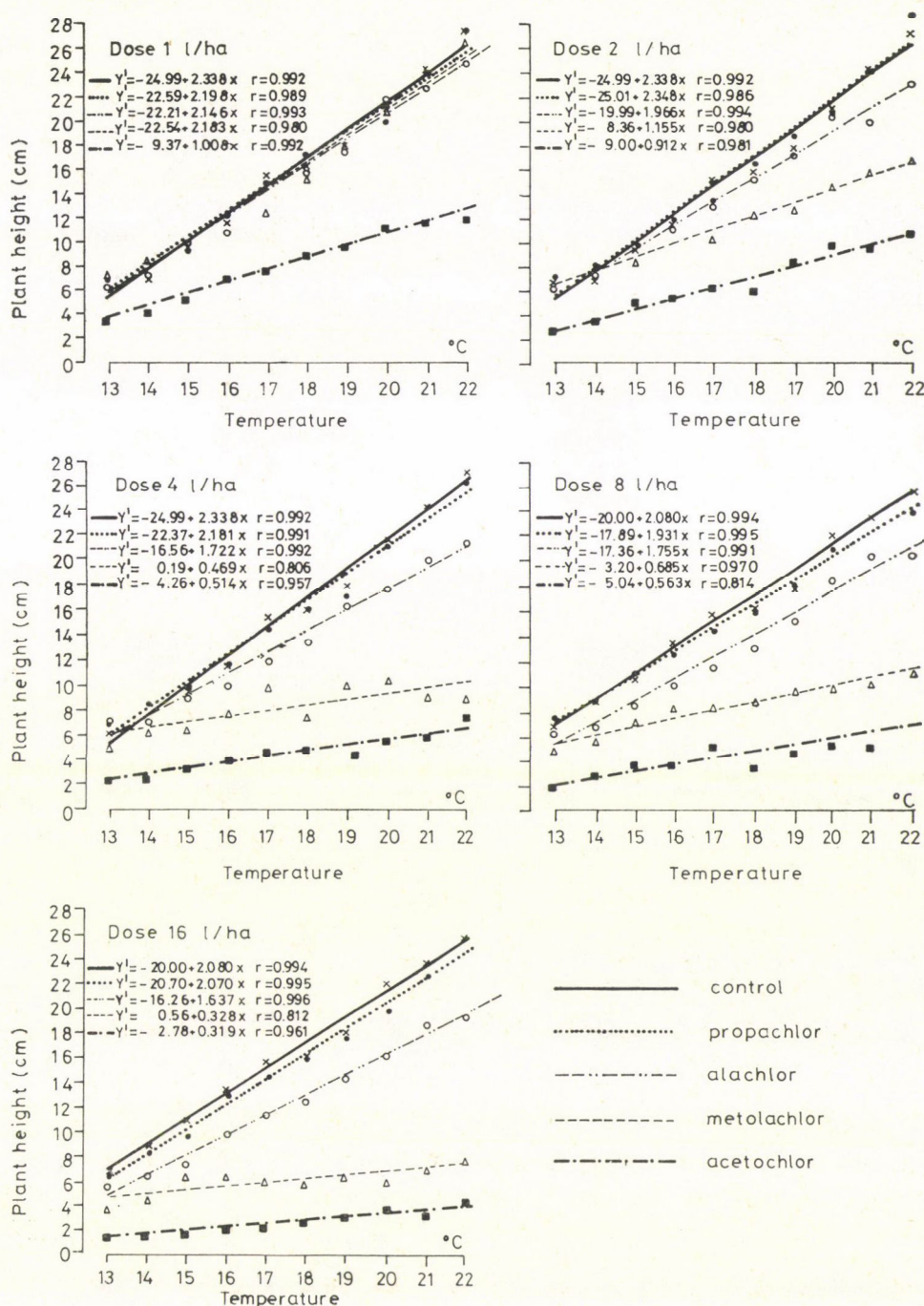


Fig. 3. Effect of different rates of the examined 4 chloroacetanilide herbicides on the height of maize plants in a gradient phytotron experiment



90.1% and 94.6%, plant height was 94.0% and 92.0%), whereas acetochlor also has a significant phytotoxic effect at this rate (the fresh weight expressed as a percentage of the control was 63.4%, plant height was 65.1%).

Raising the rates of alachlor, metolachlor and acetochlor to 2, 4 and 8 l/ha, the phytotoxic effect increased in the order listed. Regarding the degree of phytotoxic injury, the phytotoxic effect of alachlor can be considered moderate, and the curves expressing the phytotoxic effect of metolachlor and acetochlor were nearly parallel; whereas the phytotoxic effect of acetochlor exceeded that of metolachlor. This is supported by the fresh weight and plant height data expressed as a percentage of the control which, on the average of the doses used, were 78.7% and 79.7% for alachlor, 63.4% and 58.8% for metolachlor, and 38.5% and 39.6% for acetochlor.

*Comparison of the phytotoxic effect of acetanilide herbicides at temperatures between 12–22 °C and at different rates*

The effects of various rates of the 4 acetanilide herbicides tested on the shoot weight and height of maize plants as affected by a continuous linear temperature gradient between 13–22 °C are shown in Figures 2 and 3. The equations and correlation coefficients fitted to the data are indicated in the figures, the regression coefficients are summarized in Table 2. It can be seen that the (*r*) value of the correlation coefficients indicates a very close relationship, and the F-test at a *P* = 0.1% level also exhibits a significant relationship in every case.

The figures and the values of the regression coefficients reveal that the regression coefficients express the phytotoxic effect of herbicides accurately

Table 2

*“b” values expressing the changes in the fresh weight and height of maize plants as affected by different doses of the 4 chloroacetanilide herbicides studied in a gradient phytotron experiment*

Dose l/ha	Plant height (cm)				Fresh weight of shoot (g)			
	Propachlor	Alachlor	Metolachlor	Acetochlor	Propachlor	Alachlor	Metolachlor	Acetochlor
— “b” values —								
<i>Experiment 1</i>								
0	2.338	2.338	2.338	2.338	0.157	0.157	0.157	0.157
1	2.198	2.146	2.183	1.008	0.150	0.146	0.158	0.072
2	2.348	1.966	1.155	0.912	0.164	0.145	0.107	0.066
4	2.181	1.722	0.469	0.514	0.150	0.129	0.061	0.047
<i>Experiment 2</i>								
0	2.080	2.080	2.080	2.080	0.178	0.178	0.178	0.178
8	1.931	1.755	0.685	0.563	0.157	0.148	0.084	0.046
16	2.070	1.637	0.328	0.319	0.167	0.142	0.046	0.026



and are suited to compare the phytotoxic effect of herbicides in these types of experiments. A decrease in the "b" values indicates an increase of the phytotoxic effect.

Figures 2 and 3 show clearly that at 13 °C the phytotoxic effect of herbicides is less evident, as the primary limiting factor is temperature. As the temperature increases from 13 °C to 22 °C, the phytotoxic effect of the herbicides differs more and more.

Higher dosages lead to increased phytotoxicity. At the 1 l/ha rate the phytotoxic effect of acetochlor only becomes different. At the 2 l/h dose — in addition to acetochlor — the phytotoxic effect of metolachlor and alachlor becomes markedly different, too. At doses of 4 and 8 but especially of 16 l/ha three groups of the acetanilide herbicides can be distinguished regarding their phytotoxic effect. Propachlor which can be placed in the first group is not toxic at low rates and is slightly toxic at higher rates. A moderate phytotoxic injury caused by metolachlor and acetochlor in the third group is significant. At every dosage examined the phytotoxic effect, measured causing the fresh weight of the shoot and plant height as parameters, increased in the following order: propachlor, alachlor, metolachlor and acetochlor.

An analysis of the plant height and fresh weight data expressed as a percentage of the control at a given temperature did not prove — within the range examined — the phytotoxic injury increasing effect of higher temperatures. At the 4 l/ha and higher rates of metolachlor, however, phytotoxic injury increased slightly — but quite consequently — due to higher temperature. At the same time, in the case of thiocarbamate herbicides, several researchers have demonstrated that injury increases at higher temperatures (e.g. Burt and Akinsoroton 1976, Nagy and Balogh 1985).

#### *The effect of antidotes on the phytotoxic injury caused by acetochlor at identical and differing temperature conditions*

The protective effect of antidotes AD-67 and DKA-24 in various doses of acetochlor was studied in experiments set up in PGV and gradient phytotron chambers.

The effect of acetochlor and acetochlor + antidote combinations on the plant height, shoot and root weight of maize is shown in a column diagram in Figure 4. It can be seen that the examined AD-67 and DKA-24 antidotes significantly decreased the phytotoxic injury to maize plants at every rate of acetochlor. The highest antidote effect was obtained on plant height, followed by shoot and root weight. If the antidote effect is expressed as a percentage of the control and on the average of the doses applied, the following ratios are obtained: 85.7% for plant height, 80.6% for shoot weight and 80.4% for root weight.



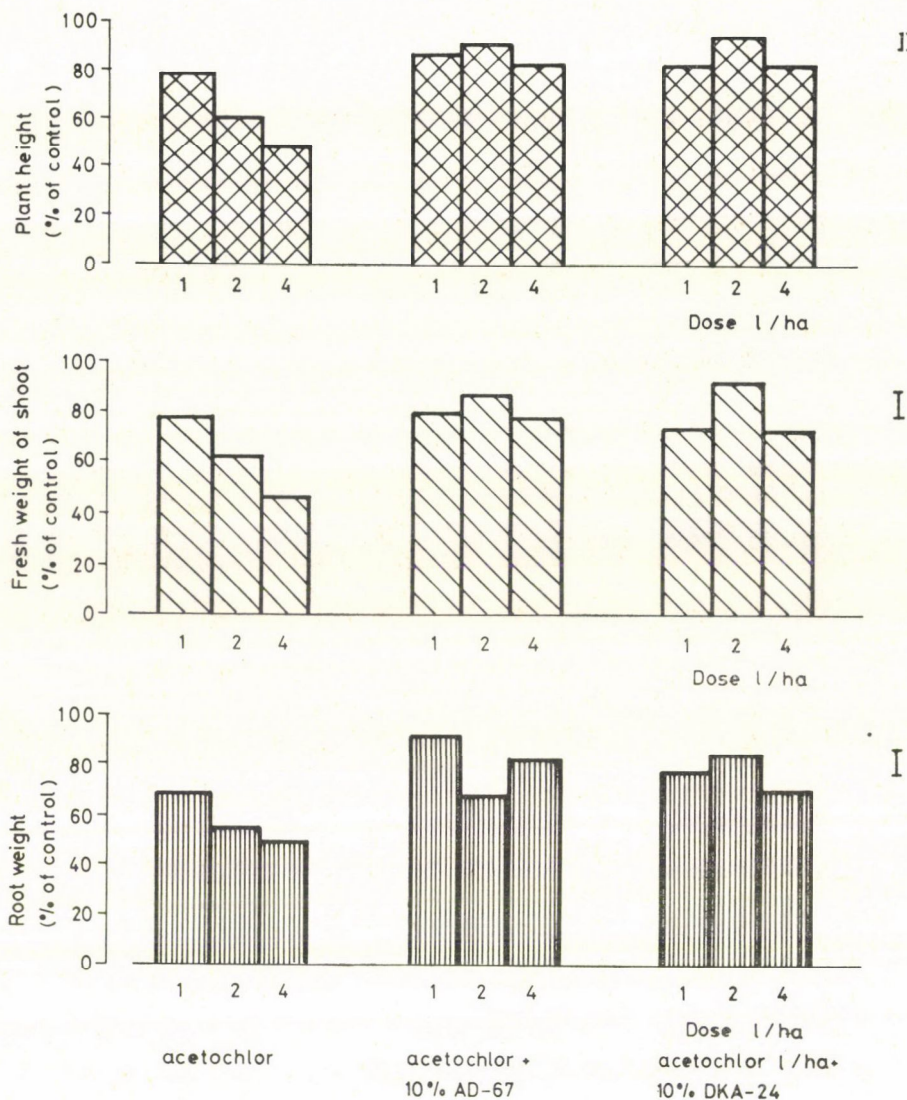


Fig. 4. Effects of antidotes AD-67 and DKA-24 at different rates of aceto-chlor

If the antidote effect is studied at different aceto-chlor dosages, then in this experiment significantly the best antidote effect is obtained at 2 l/ha aceto-chlor. There is no significant difference between the protective effect of antidotes.

Figure 5. shows the protective effect of antidotes AD-67 and DKA-24 as affected by temperature at various rates of aceto-chlor. The figure indicates the equations of the linear functions fitted to the data and the values of the

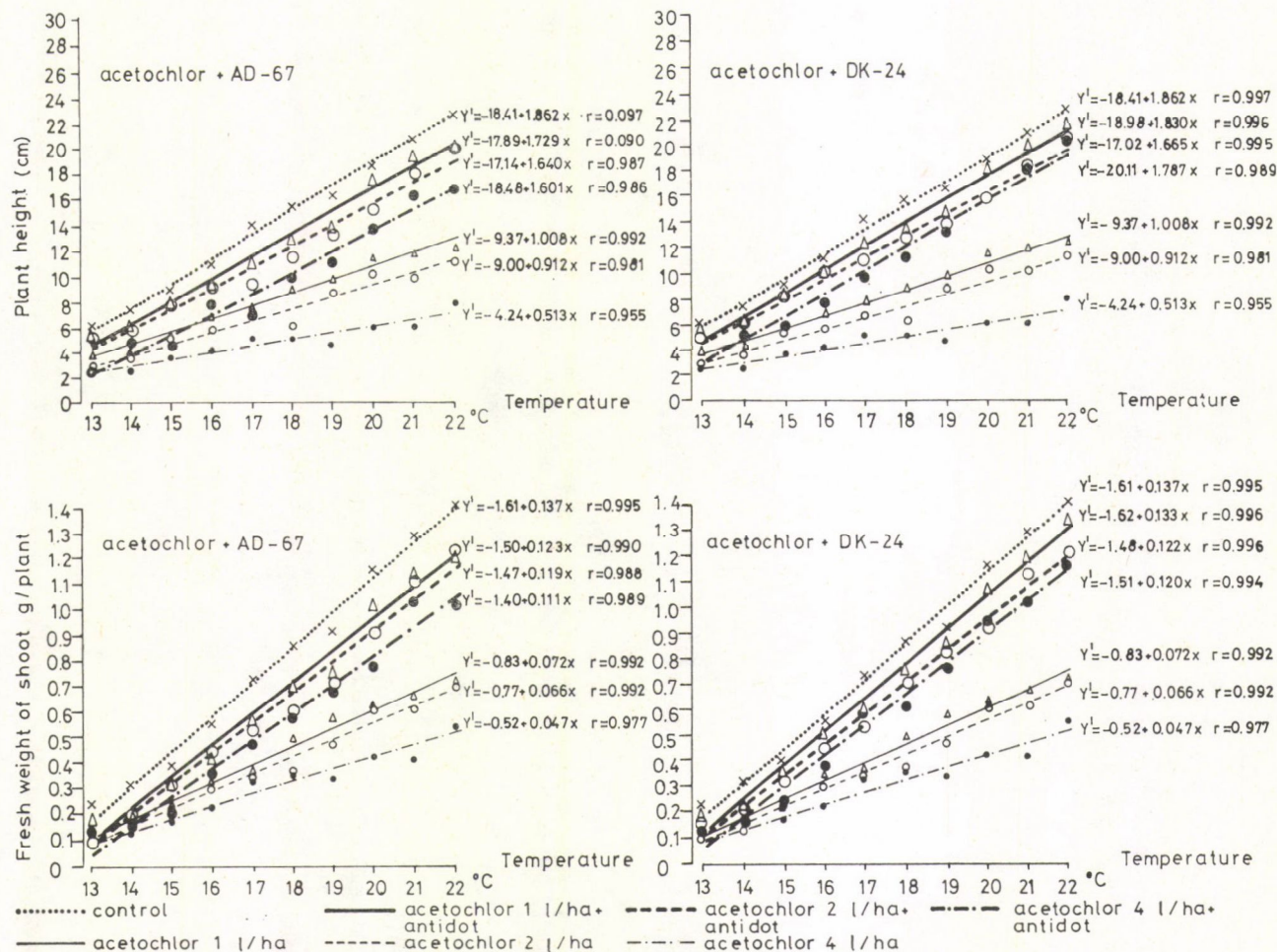


Fig. 5. Effects of antidotes AD-67 and DKA-24 on the phytotoxic injury of acetochlor in gradient phytotron experiments



Table 3

*"b" values expressing the changes in the height and shoot weight of maize plants as effected by acetochlor doses and antidotes in a gradient phytotron experiment*

Treatment	Dose		Plant height cm	Shoot weight g
	Herbicide l/ha	Antidote %		
— “b” value —				
Control			1.862	0.137
Acetochlor	1		1.008	0.072
Acetochlor+AD 67	1	10	1.729	0.123
Acetochlor+DKA-24	1	10	1.830	0.133
Acetochlor	2		0.912	0.066
Acetochlor+AD-67	2	10	1.640	0.119
Acetochlor+DKA-24	2	10	1.665	0.122
Acetochlor	4		0.513	0.047
Acetochlor+AD-67	4	10	1.601	0.111
Acetochlor+DKA-24	4	10	1.787	0.120

correlation coefficients. The relationship was significant in every case. The antidote effect was found to be good at both low and high temperatures at every acetochlor dose. It can also be seen that at higher acetochlor rates the protective effect of the antidotes does not reach the antidote effect at lower rates of acetochlor. The relationship is consequent for every parameter and antidote examined (fresh weight of shoot, plant height).

The data expressed as a percentage of the control at a given temperature show that the antidote effect is greater at higher temperatures. The interaction between temperature and the antidote effect is insignificant however. There is no significant difference between the protective effect of the antidotes.

Table 3 summarizes the regression coefficients of the linear functions fitted to the data pairs. It can be seen that the increase of the "b" values reflects the protective effect of the antidotes, whereas the decreasing "b" values indicate the increase in the phytotoxic effect of acetochlor.

### Conclusions

The consistent results of phytotron experiments arranged with maize plants at given temperature, or temperatures changing according to a linear gradient, reveal that the phytotoxic injury caused by the acetanilide herbicides studied increased in the following order:

propachlor → alachlor → metolachlor → acetochlor



The phytotoxic effect of propachlor did not increase within the dosage range examined. On the other hand a definite dosage-dependence could be observed for the herbicides alachlor, metolachlor and acetochlor.

The weight of maize shoots and the maize height are the most sensitive to the phytotoxic effect of acetanilides. Leaf area data can be less effectively used. When measuring root weight, experience revealed that it was justified to determine the shoot-root ratio (allometry). Significant differences were found between the herbicide susceptibility of hybrids in certain experiments, which in addition to the genetic background can probably be explained by differences in early vigour. In early spring, when maize is planted and acetanilide herbicides are applied, temperatures often vary between 13–22 °C. Thus, information concerning the effect of temperature on herbicide phytotoxicity can be valuable also from a practical point. The data of experiments set up in gradient phytotron chambers did not prove that the phytotoxic injury of maize plants raised at various temperatures was significantly affected by changes in temperature between 13–22 °C, according to a linear gradient. It should be mentioned, however, that the experimental data were insufficient to decide the question, in view of the fact that the maize plants were harvested at the same time at every temperature variant of the experiment. Thus in the plants grown at lower temperatures there was less dry matter accumulation than in those grown at higher temperatures. The data expressed as a percentage of the control at the same temperature lead to the conclusion that there are differences between the phytotoxicity of the herbicides tested regarding temperature, further experiments are required for the exact study of the problem in which the plants grown at different temperatures should be harvested periodically.

The experimental data of several years reveal that, compared to their phytotoxic effect, the studied herbicides show an opposite order regarding their weed control effect; i.e. acetochlor has the best weed control effect.

It should also be mentioned that in phytotron experiments favourable, provocative conditions can be created (through irrigation, temperature, soil composition, etc.) to induce the phytotoxic effect of herbicides and the comparative evaluation of the protective effect of antidotes.

Our experimental data verify that the less selective acetanilide herbicides (metolachlor and especially acetochlor) should be used in the antidoted form in order to prevent phytotoxic injury to maize plants under predisposing conditions (unfavourable weather and soil conditions, susceptible varieties). The antidotes AD-67 and DKA-24 studied in the experiments had reliable protective effect at various temperatures and different acetochlor rates.



## Acknowledgements

The authors thank Mrs. M. Kovács for her assistance in setting up and evaluating the phytotron experiments.

## References

- Burt, G. W., Akinsorotan, A. O. (1976): Factors affecting thiocarbamate injury to corn I. Temperature and soil moisture. *Weed Sci.*, **24**, 319-321.
- Chandler, J. M., Basler, E., Santelmann, P. W. (1974): Uptake and translocation of alachlor in soybean and wheat. *Weed Sci.*, **22**, 253-258.
- Chang, F. Y., Stephenson, G. R., Bandeen, J. B. (1973): N,N-diallyl-dichloroacetamide (R-25788) as an antidote for EPTC and other herbicides in corn. *Weed Res.*, **13**, 399-406.
- Dal, L. M., Hess, F. D. (1980): An analysis of the growth inhibitory characteristics of alachlor and metolachlor. *Weed Sci.*, **28**, 168-175.
- Duke, W. B., Slife, F. W., Hanson, J. B., Butler, H. S. (1975): An investigation on the mechanism of action of propachlor. *Weed Sci.*, **23**, 142-147.
- Gimesi, A. (1981): Herbicid antidótumok üvegházi és szabadföldi kísérleteinek eredménye. *Növénytermelés*, **30**, 241-248.
- Görög, K., Muschinek, Gy., Mustárdy, L. A., Faludi-Dániel, Á. (1982): Comparative studies of safeners for the prevention of EPTC injury in maize. *Weed Res.*, **22**, 27-33.
- Györfy, B. (1977): Evaluation de l'EPTC/protectant dans le maïs, 9<sup>e</sup> Conférence du Columa, Journées d'études sur les herbicides. *Compte Rendu. Paris*, **2**, 359-374.
- Hamm, Ph. C. (1974): Discovery, development, and current status of the chloroacetamide herbicides. *Weed Sci.*, **22**, 541-545.
- Jablonkai, I., Dutka, F. (1985): Metabolism of acetochlor herbicide in tolerant and sensitive plant species. *J. Radioanal. Nucl. Chem. Letters*, **94** (4), 271-280.
- Leavitt, J. R. C., Penner, D. (1978): Protection of corn (*Zea mays*) from acetanilide herbicidal injury with the antidote R-25788. *Weed Sci.*, **26**, 653-659.
- Mulder, C. E. G., Nalewaja, J. D. (1970): Temperature effect of phytotoxicity of soil-applied herbicides. *Weed Sci.*, **26**, 566-570.
- Nagy, J., Balogh, K. (1985): *A new safener for EPTC in Corn*. British Crop Protection Conference — Weeds 2-13. Brighton. 107-111.
- Pallos, F. M., Casida, J. E. (1978): *Chemistry and action of herbicide antidotes*. Academic Press, New York, San Francisco, London.
- Penner, D., Graves, D. (1972): Temperature influence on herbicide injury to navy beans. *Agron. J.*, **64**, 30.
- Rao, V. S., Duke, W. B. (1976): Effect of alachlor, propachlor, and prynachlor on GA<sub>3</sub>-induced production of protease and  $\alpha$ -amylase. *Weed Sci.*, **24**, 616-618.
- Winkle, M. E., Leavitt, J. R. C., Burnside, D. C. (1980): Acetanilide-antidote combinations for weed control in corn (*Zea mays*) and sorghum (*Sorghum bicolor*). *Weed Sci.*, **28**, 699-704.

## ON THE PRESENCE OF FUSICOCCIN IN HIGHER PLANTS

G. S. MUROMTSEV, V. D. VOBLIKOVA, N. S. KOBRINA  
V. M. KORENEVA, V. L. SADOVSKAYA and V. V. STOLPAKOVA

INSTITUTE OF AGRICULTURAL BIOTECHNOLOGY, MOSCOW, USSR

(Received 26 October 1987; accepted 8 January 1988)

The work confirms the earlier evidence from this laboratory for the presence of fusiccoccins A and C in plant of *Zea mays* L. cv. Sterling and in common head cabbage *Brassica oleracea* L. cv. Amager of the 1986 crop by HPLC combined with GC/MS et SIM.

**Keywords:** *Brassica oleracea* L. cv. Amager, cabbage, fusiccoccin contents, maize, *Zea mays* L. cv. Sterling

### Introduction

The search for fusiccoccin in higher plants stemmed from the striking analogy to the history of gibberellin, which was found in higher plants (and has since been recognized as a major phytohormone) about a quarter of a century after its isolation from a culture of a phytopathogenic fungus *Gibberella fujikuroi* (Muromtsev and Agnistikova 1984).

Fusiccoccin, like gibberellin, is a terpenoid, and was discovered in a culture of a phytopathogenic fungus, *Fusicoccum amygdali* Del. (Ballio et al. 1964); it exhibits a high and versatile physiological activity at concentrations typical of phytohormones. Like cytokinin, fusiccoccin is antagonistic to abscisic acid in opening the stomata and promoting seed germination (Marre 1979). Like auxin, fusiccoccin evokes root formation in cuttings (Sultonov and Muromtsev 1985). Of particular importance is the finding of specific protein receptors for fusiccoccin in higher plants (Pesci et al. 1979).

The only attempt to detect fusiccoccin in higher plants of which we are aware was unsuccessful: in 1976 a group of Italian researchers reported that fusiccoccin was absent from the healthy fruits of peach and almond though observable in tissues infected with *Fusicoccum amygdali* (Ballio et al. 1976).

Preliminary positive results of the search along this line were first published by us in 1980 (Muromtsev et al. 1980). These results could not be reproduced by the Italian group (Aducci et al. 1985). Since then we have essentially revised and improved the technique for detecting fusiccoccin in plant material. The results of its application to identify fusiccoccins A and C in roots and immature cobs of *Zea mays* L. cv. Sterling and in headed cabbage



*Brassica oleracea* L. cv. Zimovka 1474 of the 1985 crop have been published elsewhere (Muromtsev et al. 1986, and Muromtsev 1986).

Here we confirm the presence of endogenous fusicoccin in the kernel of *Zea mays* L. cv. Sterling and in *Brassica oleracea* L. cv. Amager of the 1986 crop.

### Materials and methods

#### Maize kernel extraction and purification for HPLC

Ears of maize cv. Sterling were harvested in August 1986 at milky-way ripeness. The kernels (5 kg) were separated, ground, and extracted with acetone (1 : 2) for 4 weeks. The water-acetone extract was evaporated under reduced pressure to an aqueous residue (2.81) which was then twice extracted with chloroform (1 : 1 and 1 : 0.5) by shaking for 30 min. The chloroform extract was dehydrated with  $\text{Na}_2\text{SO}_4$  and evaporated under reduced pressure to a solid residue (11.8 g) which was washed with 120 ml hexane, the solution discarded, and the solid residue (1.06 g) dissolved in 20 ml  $\text{MeOH-H}_2\text{O}$  (60 : 40). The filtered solution was applied onto the HPLC column. The dissolved substance in the sample weighed 0.43 g.

#### Cabbage leaves extraction and purification for HPLC

Common head cabbage cv. Amager was harvested in September 1986; the leaves (7 kg) were detached, ground, and extracted with acetone (1 : 1.5) for one month. The water-acetone extract was evaporated under reduced pressure to an aqueous residue (3.6 l) and extracted twice with chloroform (1 : 1 and 1 : 0.5). The combined extracts were dehydrated with  $\text{Na}_2\text{SO}_4$  and evaporated to dryness under reduced pressure. The residue (4.65 g) was washed with 100 ml hexane and the remaining oil (0.94 g) was dissolved in 10 ml methanol. Methanol was removed under reduced pressure and the residue (0.87 g) dissolved in 10 ml of  $\text{MeOH-H}_2\text{O}$  (60 : 40). The filtered solution was applied onto the HPLC column. The sample contained 0.33 g of dissolved substance.

#### High performance liquid chromatography

A Du Pont model 8800 liquid chromatograph was used for HPLC; samples were introduced with a Rheodyne 7125 injector with a 2 ml loop. Reversed-phase HPLC was performed on  $9.4 \times 250$  mm Zorbax  $\text{C}_8$  columns with  $\text{MeOH-H}_2\text{O}$  and  $\text{MeCN-H}_2\text{O}$  systems; elution was monitored with a UV detector at 230 nm. Samples were purified in several consecutive runs, by collecting and recycling fractions with retention times coincident with those of authentic fusicoccins A and C. First run:  $\text{MeOH-H}_2\text{O}$ , (65 : 35), 35 °C, 3.5 ml/min; second run:  $\text{MeOH-H}_2\text{O}$  (60 : 40), 50 °C, 4 ml/min; third run  $\text{MeCN-H}_2\text{O}$  (60 : 40), 50 °C, 3 ml/min. If adequate purity was not attained, the fractions were further rechromatographed under the conditions of the second and third runs.

#### Derivatization

The trimethylsilyl derivatives were obtained by mixing the dry residue of the analysed substance with 10  $\mu\text{l}$  of the TMSi-S-Universal reagent (Serva) consisting of BSTFA : TMCS : TSIM (3 : 2 : 3) in 40  $\mu\text{l}$  abs. benzene and incubating the mixture for 30 min at 20 °C.

#### GC/MS

A Hitachi M-80 A mass spectrometer was equipped with a M-003 computerized data processing system. The column was 2% OV-1 on Gas Chrom Q 100-120 mesh (1 m  $\times$  3 mm i.d.); column temp. 280 °C with preheating from 240 °C to 280 °C at 10 °C/min; injector and separator temp. 250 °C; He carrier gas flow, 35 ml/min. Ionization, EI (70 eV); ionization chamber temp. 200 °C.



### Results and discussion

In the plant, the presence of fusicoccin can be assumed both in free and bound state. The aim of the present study was to detect free fusicoccin.

The plant material was ground and extracted with acetone at room temperature for 10–30 days without stirring. The acetone extract was evaporated under reduced pressure to an aqueous residue which was then extracted with chloroform. The chloroform extract was dehydrated and vacuum-evaporated; samples were fractionated by 3–4 cycles of reversed-phase semipreparative HPLC on Zorbax C<sub>8</sub> columns in MeOH–H<sub>2</sub>O and MeCN–H<sub>2</sub>O systems.

Chromatographic conditions were designed with the use of solutions of authentic fusicoccin and plant extracts with added exogenous fusicoccin. The presence of plant admixtures was found not to affect the retention time of fusicoccin on HPLC columns.

The HPLC fraction with the retention time identical to that of authentic fusicoccin (on the same column under the same conditions) was collected and silylated. The mixture of trimethylsilyl (TMS) derivatives was subjected to GC/SIM analysis in the electron impact mode.

We have previously (Sadovskaya et al. 1986) proposed a method of identifying fusicoccin metabolites in the culture medium of *Fusicoccum amygdali* by GC/MS of TMS derivatives. The cleavage of the TMS derivatives of fusicoccin metabolites was studied with tetra-TMS fusicoccin A (I), the molecular ion of which is split under the electron impact into several characteristic fragments shown in the scheme (Fig. 1).

Obviously, the splitting of the glycoside link in the molecular ion yields aglycone fragments A and B ( $m/z$  535 and 551, respectively) whereas the cleavage of C–C bonds 6–7, 1–11, and 8–9 of the eight-membered ring both in the molecular ion  $M^+$  and in the aglycone fragment A gives rise to ions

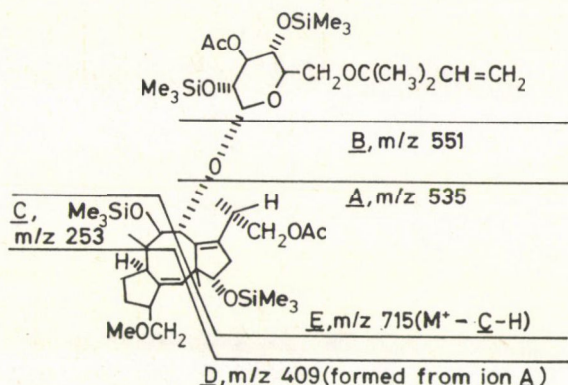


Fig. 1. The fragmentation patterns of tetra-TMS fusicoccin A (I) under electron impact



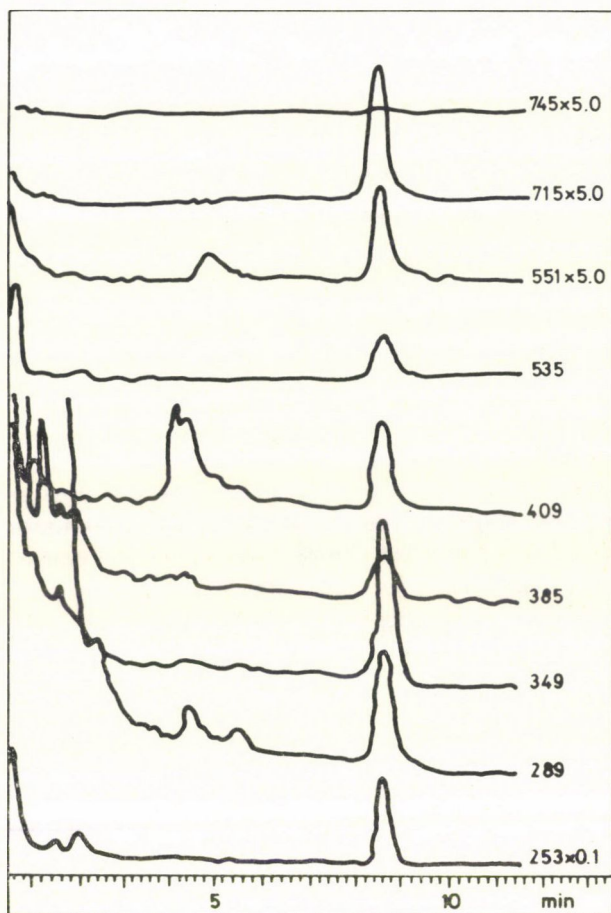


Fig. 2. GC/SIM profiles identifying tetra-TMS fusicoccin A in the HPLC fraction of the maize kernel extract

*C*, *D*, and *E* with  $m/z$ , of 253, 409 and 715 in the mass spectrum of *I*. In addition, this spectrum also exhibits peaks of ions pertaining to the glucopyranose moiety with  $m/z$  349 and 289.

These ions *A*–*E* and the  $m/z$  385, 349 and 289 ions were chosen as markers for detecting endogenous fusicoccin A in the plant material by selected ion monitoring.

Figures 2 and 3 present to GC/SIM chromatograms identifying fusicoccin A in maize kernel and headed cabbage of the 1986 crop. As can be seen, the GC/SIM profiles of the HPLC fractions under study proved to be identical to those of the tetra-TMS fusicoccin A standard both in the retention time of the peaks of  $m/z$  253, 349, 385, 409, 535 and 715 and in their relative inten-

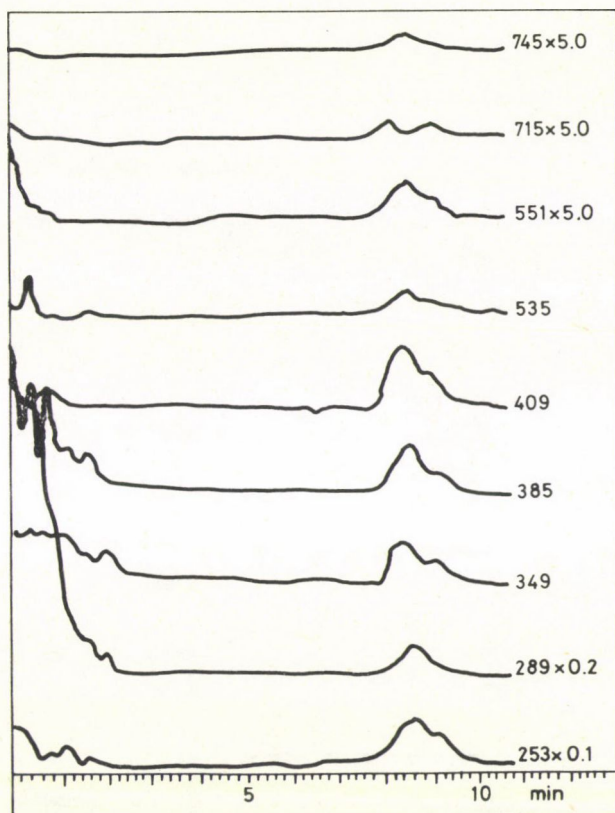


Fig. 3. GC/SIM profiles identifying tetra-TMS fusicoccin A in the HPLC fraction of the cabbage leaf extract

sities (Fig. 4). Also, a partial mass spectrum of the trimethylsilylated HPLC fraction of the cabbage leaf extract recorded by GC/MS in the 200–800 amu region was similar to that of authentic tetra-TMS fusicoccin A under the same conditions (Fig. 5).

Thus, the combination of chromatographic and mass spectrometric data provide convincing evidence for the presence of fusicoccin A in maize kernel and headed cabbage.

Solutions of the fusicoccin A standard were found to gradually from small amounts of fusicoccin C which could be separated by HPLC. During preparative HPLC of cabbage leaf and maize kernel extracts, a fraction was also collected with retention time corresponding to fusicoccin C. The GC/SIM (Figs 6a, b, c) of the trimethylsilyl derivatives of this fraction and authentic fusicoccin C (with a minor admixture of fusicoccin A) turned out to be quite similar, both exhibiting peaks at  $m/z$  535, 551 and 745 (the *E*-type ion) char-



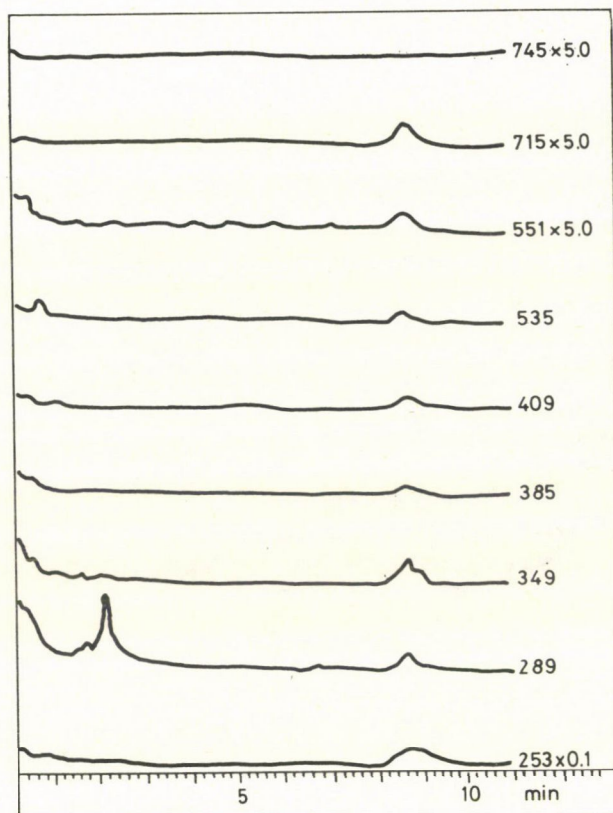


Fig. 4. GC/SIM profiles of authentic tetra-TMS fusicoccin A, 5ng

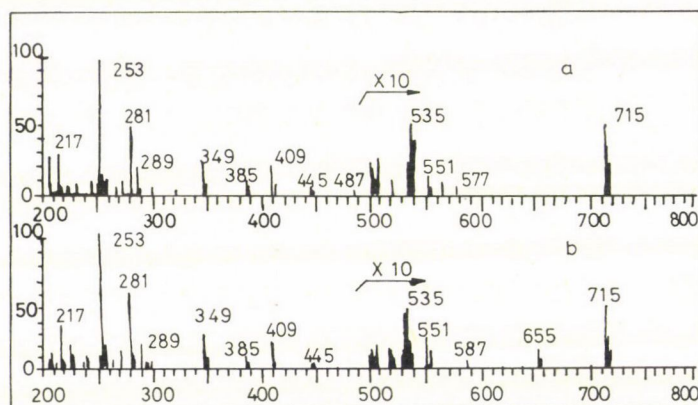


Fig. 5. Partial mass spectra of (a) tetra-TMS fusicoccin A in the HPLC fraction of the cabbage leaf extract and (b) authentic tetra-TMS fusicoccin A

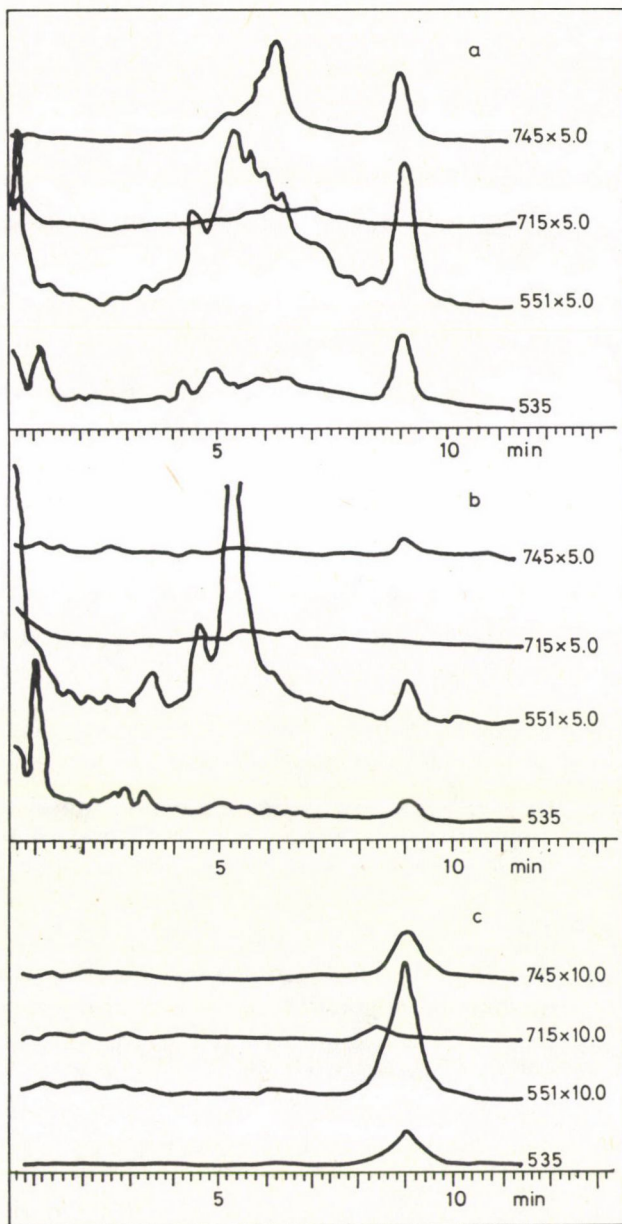


Fig. 6. GC/SIM profiles identifying penta-TMS fusicoccin C (a) in the HPLC fraction of the cabbage leaf extract; (b) in the HPLC fraction of the maize kernel extract; (c) in the standard solution



acteristic of penta-TMS fusicoccin C, and thus indicating that the extracts contained fusicoccin C as well. It can not, however, be ruled out that fusicoccin C could have been formed from fusicoccin A during isolation and chromatography, since conversion of fusicoccin A into fusicoccin C is a known fact (Ballio et al. 1977).

Because the work involved the use of a standard solution of fusicoccin A, to ensure the reliability of the analysis all of the equipment was thoroughly checked for the absence of fusicoccin.

Thus, our effort made it possible to detect fusicoccin A in amounts about  $10^{-8}$  g/kg fresh wt of cabbage leaves or maize kernel.

In our experience, the crucial points in identifying fusicoccins, which are present in the plants at extremely low concentrations, are the sample purity and the detection technique employed. The use of HPLC permitting preparative isolation of fusicoccin-enriched fractions, and of mass spectrometry to identify fusicoccin in these fractions, determined the final success of the analysis.

It must be emphasized that the GC/SIM technique used here to detect fusicoccins is no less sensitive than RIA (the lower detection limit for isolated fusicoccin is several picomoles) and probably even more selective, especially if advantage can be taken of ions with high mass numbers (e.g. the  $m/z$  715 ion for fusicoccin A and  $m/z$  745 ion for fusicoccin C).

The described technique for detecting fusicoccins in higher plants was employed with moderate amounts of plant material (1–7 kg). For substantially larger amounts (50–150 kg) the chloroform extract residue prior to HPLC was additionally purified on a silica gel column, collecting the fraction eluted with chloroform-isopropanol (9 : 1) (Muromtsev et al. in press).

Of course, the above results should at present be treated with some caution, since there still remains a possibility that the fusicoccin had been produced not by the plants per se but by some contaminating microorganisms. The same kind of argument is applicable to other phytohormones known to be synthesized by certain microorganisms (and by diverse ones as in the case of auxins and ethylene), and generally to a vast number of compounds found in plants in very small amounts. Here such contamination appears to be highly improbable because fusicoccin is only known to be made by a scantily occurring phytopathogen *Fusicoccum amygdali* absent from the USSR.

In any case the very fact of the presence of fusicoccin in higher plants is new and undoubtedly worth attention in itself. Studies including experiments under sterile conditions are under way to ascertain its endogenous origin.

## References

- Aducci, P., Ballio, A., Evidente, A., Federico, R., Iasiello, I., Marra, M., Randazzo, G. (1985): *Phytochemistry*, **24**, 1097.
- Ballio, A., Chain, E. B., De Leo, P., Erlanger, B. F., Mauri, M., Tonolo, A. (1964): *Nature (London)* **203**, 297.
- Ballio, A., d'Alessio, V., Randazzo, G., Bottalico, A., Graniti, A., Sparapano, L., Bosnar, B., Casinovi, C. G., Griбанovski-Sassu, O. (1976): *Physiol. Plant. Path.* **8**, 163.
- Ballio, A., Federico, R., Scalorbi, D. (1977): *Rend. Accad. Naz. Lincei* **63**, 604.
- Marre, E. (1979): *Ann. Rev. Plant. Physiol.* **30**, 273.
- Muromtsev, G. S., Kobrina, N. S., Voblikova, V. D., Koreneva, V. M. (1980): *Izv. AN SSSR. Ser. Biol. No. 6*, 897.
- Muromtsev, G. S., Agnistikova, V. N. (1984): *Gibberellins*. Nauka, Moscow.
- Muromtsev, G. S., Voblikova, V. D., Kobrina, N. S., Koreneva, V. M., Sadovskaya, V. L., Stolapkova, V. V. (1986): *Dokl. Vaskhnil No. 3*, 2.





## *Animal production and genetics*

### COMPOSITION OF COLOSTRUM FROM GOATS AND EWES DROPPING TWINS

J. CSAPÓ, GY. WOLF and Zs. CSAPÓ

UNIVERSITY OF AGRICULTURE, FACULTY OF ANIMAL SCIENCE  
KAPOSVÁR, HUNGARY

(Received: 16 April 1987; accepted in revised form 23 November 1987)

The authors determined the colostrum of 11 single and 7 twin-lambing Hungarian combing merino ewes and 6 single- and 4 twin-dropping improved Hungarian white goats for dry matter-, total protein-, true protein-, whey protein, true whey protein-, immunoglobulin-G-, casein- and NPN content as a function of the postpartum time. Furthermore, they analysed the colostrum and colostral protein for amino acid composition, biological value, macro- and microelement content.

They found that the first milked colostrum of the twin-dropping ewes and goats contained significantly more dry matter, total protein, true protein, whey protein, true whey protein and immunoglobulin-G than that of mothers with a single progeny. In the other components examined there was no significant difference between the two groups. Twenty-four hours after parturition the mentioned differences in the composition of colostrum disappeared and could not even be detected subsequently in a single case.

**Keywords:** biological value of colostrum, colostrum composition, goats, immunoglobulin-G and protein content of colostrum, macro- and microelement content sheep/ewes, single-dropping, twin-dropping

#### Introduction

In earlier studies when analysing the colostrum, postcolostral milk and milk of various genotypes of cattle (Csapó et al. 1982a, 1982b, Csapó and Csapó-Kiss 1984a), goat (Csapó et al. 1987) and ewe (Csapó et al. 1986a) we noticed that the colostrum of dams dropping twins differed somewhat in composition from the colostrum of those with a single progeny. The difference which was considerable, immediately after dropping soon disappeared. Since at that time colostrum was available for us from only a few dams with twins, we did not publish our results.

In 1985 and 1986 we had an opportunity to analyse the colostrum and milk of ewes and goats dropping twins and compare them with the colostrum of dams with a single progeny. In the study 7 twin-dropping Hungarian combing merino ewes and 4 twin-dropping Hungarian white she-goats crossed with boer he-goats were included. The present paper describes the results of these two years of work.



Before determining the composition of cow's, goat's and ewe's colostrum, we read through the relevant foreign and Hungarian literature, but did not find any reference to whether twin-dropping had an influence on the composition of the colostrum. Since the aim of our present work is to give a comparative evaluation of the colostrum of dams dropping singles and twins, respectively, we do not deem it necessary to present literary data on goat's and ewe's colostrum, since single- or twin-dropping is not indicated. Consequently, this paper contains no literary review.

## Materials and methods

### *Goats*

The experiments were carried out with 10 improved Hungarian white goats derived at Szigetcsép from crossing with boer he-goats obtained from the German Federal Republic, 6 of which dropped one each and 4 dropped twins. The breed is characterized by white colour, short hair, absence of horn and excellent milking capacity. The annual milk production of the ewes was 700–800 kg on the average of 300 days of lactation; the height of withers of the mature mother goats was 70–80 cm, and their weight 50–80 kg. In the course of the experiment the mother goats were kept in goat-folds under free-pen condition in groups of 30, with 3 m<sup>2</sup> area for each mother goat. The analyses were made of the colostrum of mother goats dropping between 10 February and 10 March 1986. Dropping took place in single boxes where the kid was left with its mother for 3 days after birth, then on the 4th day both were transferred to the kid-pen. Immediately after dropping, then between the 20th and 28th hour, about the 48th hour and on the 3rd day after dropping colostrum, on the 5th day after dropping postcolostral milk- and on the 7th day, milk samples were taken. The colostrum samples (about 50–100 cm<sup>3</sup>) were extracted by hand, the milk samples were collected by means of an ALFA-LAVAL type milking machine. During the experiment the animals consumed wheat-, barley-, pea- and vetch straw, and good quality maize silage ad libitum, occasionally supplemented with fodder cabbage, meadow- and leguminous hay.

### *Sheep*

The 18 Hungarian combing merino ewes examined — of which 11 dropped one lamb each and 7 lambed twins — were so chosen from a flock of 450 reared on the Experimental Grounds of the Keszthely University of Agricultural Sciences, Faculty of Animal Sciences, Kaposvár as to form a representative sample as regards the major parameters. The experiment was carried out with ewes lambing between 15 May and 15 July 1985. Colostrum samples were taken immediately after lambing, then between the 20th and 28th hour, and on the third day after lambing; postcolostral samples were taken on the 4th to 5th day, and milk samples on the 10th, 20th and 30th days of lactation. Sampling was made by hand in each case. During the experimental period the animals were turned out to pasture and in addition consumed about 0.4 kg/ewe/day of a mixture of farm grains.

### *Chemical analysis of the samples*

The milk samples were filtered through gauze, then stored at  $-20^{\circ}\text{C}$  until processed. The dry matter content was determined according to the standard MSZ 3744-67, by exsiccation to constant weight.

The protein contents and protein fractions of the samples were determined with a "Kjel-Foss 16200" type quick nitrogen analyser. The protein fractions of the milk were separated as follows: butter fat was removed from the whole milk by centrifuging for 10 minutes at 8000 r.m., then the value of pH was adjusted to 4.55 by means of a "OP-264" type pH-meter. The precipitated casein was removed from the whey by centrifuging for 10 minutes at 8000 r.m. The whey protein content ( $\text{N} \times 6.38$ ) after precipitated with 12.5% trichloroacetic acid was removed from the whey, then the clear pure solution was determined for nitrogen content (NPN). With NPN subtracted from the nitrogen content of the whole milk the true

protein nitrogen content of milk, and when it was subtracted from the nitrogen of the whey protein the true whey protein nitrogen content of milk was obtained. The nitrogen content of casein was obtained by deducting the nitrogen content of whey from the total nitrogen content. The nitrogen content of the fractions concerned when multiplied by the conversion factors 6.38 gave the protein content.

The immunoglobulin-G content of the colostrum was determined in the central laboratory of the Institute with "The single radial immuno-diffusion method" described by Mancini et al. (1965). The measuring results were checked in the Out-Patients' Department of Somogy-country's Council Hospital. The anti-goat Ig G rabbit serum (3C-RbAGt-Ig G) and anti-sheep Ig G rabbit serum (3C-RbASh-Ig G) as well as the goat and sheep Ig G standards were obtained from the Gödöllő- and Budapest units of the Human Serum Production- and Research Institute.

The macro- and microelement contents of the colostrum and milk were determined as described in the publication of Csapó and Csapó-Kiss (1984b), while the determination of the amino acid composition was carried out by the method of Moore and Stein (1951) according to the description of Csapó and Csapó-Kiss (1986b). The biological value was calculated on the basis of the amino acid composition, by the method of Morup and Olesen (1976).

#### Statistical analysis of the results

The mean value and scatter of the results were calculated and the mean values compared with single-aspect variance analysis by means of a HT-PTK-1096 pocket computer.

**Table 1**  
*Changes in the dry matter content and protein fractions of ewe's colostrum in response to twin-lambing*

Component, %	Time after lambing (hours)								
	0.5-1.5		$\bar{d}$	22-26		$\bar{d}$	44-52		$\bar{d}$
	single	twin		single	twin		single	twin	
	lambing			lambing			lambing		
n	11	7		11	7		11	7	
Dry matter	26.51	30.28	*	22.11	22.88	Ø	19.64	19.62	Ø
± s	2.12	1.78		2.43	2.29		1.83	1.79	
Total protein	16.98	20.14	*	9.72	10.40	Ø	6.28	6.36	Ø
± s	1.63	1.82		1.12	1.23		10.1	1.05	
True protein	16.58	19.71	**	9.36	10.02	Ø	6.03	6.11	Ø
± s	1.54	1.70		1.06	1.16		0.094	0.097	
Whey protein	11.08	14.81	**	6.38	6.52	Ø	3.31	3.36	Ø
± s	0.99	1.03		0.51	0.54		0.049	0.047	
True whey protein	10.68	14.38	**	6.02	6.14	Ø	3.06	3.11	Ø
± s	0.84	0.97		0.43	0.47		0.046	0.045	
Casein	5.89	5.63	Ø	3.34	3.88	Ø	2.97	3.00	Ø
± s	0.49	0.53		0.29	0.32		0.33	0.31	
NPN x 6.38	0.401	0.432	Ø	0.362	0.385	Ø	0.251	0.253	Ø
± s	0.043	0.042		0.041	0.040		0.039	0.038	

\*  $P < 1\%$   
 \*\*  $P < 0.1\%$   
 Ø  $P > 10\%$



## Results

Changes in the dry matter content and protein fractions of colostrum from ewes dropping singles and twins, respectively, as a function of the time passed after lambing are shown in Table 1, while the changes of the goat's colostrum are contained in Table 2. In Table 3 changes in the immunoglobulin-G content of the colostrum of ewes and goats dropping singles and twins, respectively, are seen. Table 4 shows the biological value of the ewe's and goat's colostrum calculated on the basis of the amino acid composition, while Table 5. contains the changes of macro- and microelements.

An analysis of the data of Tables 1 and 2 reveals that immediately after parturition the colostrum of goats dropping twins contains about 5.9% more dry matter, 3.4% more total protein, 3.7% more whey protein and true whey protein, 0.5% more casein and 0.06% more calculated protein ( $\text{NPN} \times 6.38$ ) than the colostrum of those dropping a single kid. The differences in dry matter and total protein are significant at  $P = 1\%$ , while in true protein,

**Table 2**

*Changes in the dry matter content and protein fractions of goat's colostrum in response to twin-dropping*

Component, %	Time after dropping (hours)								
	0.5-1.5		$\bar{d}$	22-26		$\bar{d}$	44-52		$\bar{d}$
	single	twin		single	twin		single	twin	
	dropping			dropping			dropping		
n	6	4		6	4		6	4	
Dry matter	26.38	32.25	*	19.81	20.45	Ø	18.08	18.20	Ø
± s	2.98	3.21		2.62	2.51		1.65	1.58	
Total protein	15.28	18.65	*	7.50	7.73	Ø	5.94	5.99	Ø
± s	1.83	1.92		1.43	1.52		0.98	0.99	
True protein	14.92	18.20	**	7.28	7.51	Ø	5.75	5.80	Ø
± s	1.54	1.67		1.34	1.38		0.84	0.77	
Whey protein	9.01	12.68	**	3.13	3.28	Ø	1.83	1.81	Ø
± s	1.02	1.10		0.65	0.69		0.54	0.51	
True whey protein	8.58	12.26	**	2.93	3.06	Ø	1.63	1.62	Ø
± s	0.83	0.88		0.62	0.57		0.49	0.50	
Casein	5.61	6.08	Ø	4.41	4.38	Ø	4.17	4.12	
± s	0.51	0.48		0.38	0.41		0.32	0.35	
NPN x 6.38	0.358	0.420	Ø	0.211	0.221	Ø	0.182	0.179	Ø
± s	0.069	0.067		0.051	0.062		0.049	0.047	

\*  $P < 1\%$

\*\*  $P < 0.1\%$

Ø  $P > 10\%$

Table 3

*Changes in the immunoglobulin-G content of ewe's and goat's colostrum in response to twin-dropping (mg/dm<sup>3</sup>)*

Species	Time after dropping (hours)								
	0.5-1.5		$\bar{d}$	22-26		$\bar{d}$	44-52		$\bar{d}$
	single	twin		single	twin		single	twin	
	dropping			dropping			dropping		
Ewe									
n	11	7		11	7		11	7	
$\bar{x}$	98.7	118.4	**	22.4	23.1	Ø	6.1	5.8	Ø
$\pm s$	9.8	12.1		6.5	8.1		2.2	2.4	
Goat									
n	1	4		6	4		6	4	
$\bar{x}$	112.2	132.4	**	24.1	24.8	Ø	5.9	6.3	Ø
$\pm s$	8.7	11.2		5.9	7.2		2.9	2.1	

\*\*  $P < 1\%$   
 Ø  $P > 10\%$

wey protein and true wey protein at  $P = 0.1\%$ . As regards casein- and calculated protein ( $\text{NPN} \times 6.38$ ) content significant difference between the two groups could not be pointed out. Twenty-four and forty-eight hours after parturition these differences disappear, and significant difference in the composition of colostrum can no longer be detected.

The analysis of the ewe's colostrum led to the same conclusion. Immediately after parturition the colostrum of ewe's lambing twins contained 3.8% more dry matter, 3.2% more total protein and true protein, 3.7% more wey protein and true wey protein, 0.06% more  $\text{NPN} \times 6.38$  and 0.26% less casein than the colostrum of those dropping one lamb. The differences were significant at  $P = 1\%$  for the dry matter- and total protein content, and at 0.1% for the true protein-, wey protein- and true wey protein content. As for the casein- and  $\text{NPN} \times 6.38$  content the two groups — like those of the goats — were found to have the same composition of colostrum. Twenty-four and forty-eight hours later the differences — as in the case of goats — disappeared, and from that time on ewes dropping one and two lambs, respectively showed the same composition of colostrum.

The ewe's colostrum extracted first contains 19.7 mg/kg and the goat's colostrum extracted first 20.2 mg/kg more immunoglobulin-G in the case of dams dropping twins compared to those with a single progeny. The difference of both species is significant at  $P = 0.1\%$ . Twenty-four hours after parturition no difference in the immunoglobulin-G content of colostrum between the two groups of either species could be pointed out (Table 3).



The biological value of the colostrum, both immediately after parturition and 24 hours later, is higher with goats and ewes dropping twins than with those giving birth to a single progeny. This can supposedly be explained by the higher ratio of whey protein in the colostrum after twin-birth, as it is well known that the biological value of the whey protein is higher than that of the casein. The reliability of differences in mean value cannot be proved by statistical analysis. Moreover, 48 hours after parturition even the mean values are practically identical (Table 4).

The analysis of the data in Table 5. indicates difference in the macro- and microelement content of colostrum between dams with one progeny and those with twins.

Altogether it can be established that in the case of either ewes or goats the colostrum of first milking of dams dropping twins contains significantly more dry matter, total protein, true protein, whey protein, true whey protein and immunoglobulin-G than that of dams giving birth to a single progeny. In milk samples collected later this difference cannot be pointed out. In other cases significant difference in the casein-, NPN $\times$ 6.38-, macro- and microelement content and in the biological value could not be detected even in the first milked colostrum, despite occasional differences in the mean values.

Since the immunoglobulin-G is a part of the whey protein, and the latter in turn is a part of the total protein content, it seems that the differences found in the colostrum of first milking are due primarily to an immunoglobulin-G or whey protein surplus. Hence it follows that since the quantity

**Table 4**  
*Biological value of ewe's and goat's colostrum in the case of  
single- and twin-dropping*

Species	Time after dropping (hours)								
	0.5-1.5			22-26			44-52		
	single	twin	$\bar{d}$	single	twin	$\bar{d}$	single	twin	$\bar{d}$
	dropping			dropping			dropping		
Ewe									
n	11	7		11	7		11	7	
$\bar{x}$	108.55	115.62	$\emptyset$	101.24	112.45	$\emptyset$	89.99	90.63	$\emptyset$
$\pm s$	11.71	12.38		12.35	11.25		9.13	7.63	
Goat									
n	6	4		6	4		6	4	
$\bar{x}$	127.44	137.76	$\emptyset$	113.85	124.43	$\emptyset$	110.39	110.17	$\emptyset$
$\pm s$	12.85	10.79		13.74	11.22		11.05	8.45	

$\emptyset = P > 10\%$

Table 5

*Macro- and microelement contents of ewe's and goat's colostrum  
in the case of single- and twin-dropping (mg/kg)*

Species	Time after dropping (hours)								
	0.5-1.5			22-26			44-52		
	single	twin	$\bar{d}$	single	twin	$\bar{d}$	single	twin	$\bar{d}$
	dropping			dropping			dropping		
Ewe									
n	11	7		11	7		11	7	
Potassium	1481	1503	Ø	1352	1384	Ø	1341	1329	Ø
Sodium	949	967	Ø	699	674	Ø	670	675	Ø
Calcium	2437	2398	Ø	1977	1982	Ø	1889	1894	Ø
Phosphorus	1829	1842	Ø	1538	1497	Ø	1482	1439	Ø
Magnesium	235	241	Ø	163	174	Ø	148	142	Ø
Manganese	0.302	0.315	Ø	0.167	0.172	Ø	0.143	0.149	Ø
Goat									
n	6	4		6	4		6	4	
Potassium	1514	1602	Ø	1516	1524	Ø	1539	1562	Ø
Sodium	601	684	Ø	485	497	Ø	433	496	Ø
Calcium	2303	2330	Ø	1933	1862	Ø	1815	1901	Ø
Phosphorus	2095	2002	Ø	1576	1491	Ø	1380	1415	Ø
Magnesium	294	289	Ø	167	173	Ø	125	129	Ø
Manganese	0.119	0.123	Ø	0.095	0.097	Ø	0.081	0.079	Ø

Ø = P > 10%

of first milked colostrum is limited, the mother by increasing the concentration of immunoglobulins in the same volume of milk — tries to satisfy the requirements of twins and develop a passive immunity in them.

### Summary

The authors determined the dry matter-, total protein-, true protein-, whey protein-, true whey protein-, immunoglobulin-G-, casein- and NPN content in the colostrum of 11 single-lambing- and 7 twin-lambing Hungarian combing merino ewes and 6 single-dropping and 4 twin-dropping Hungarian white goats improved with boer he-goats, as a function of the post-partum time. In addition, they analysed the colostrum and colostrum protein for amino acid composition, biological value, macro- (potassium, sodium, calcium, phosphorus, magnesium) and microelement (zinc, iron, copper, manganese) content.

The authors found that the first colostrum (milked 0.5-1 hour after parturition) of ewes and goats dropping twins contained significantly (P =



= 0.1–1.0%) more dry matter and total protein and significantly  $P = 0.1\%$ ) more true protein, whey protein, true whey protein and immunoglobulin-G than that of dams with a single progeny. As regards the casein- and NPN content, the amino acid composition and the macro- and microelement content, no significant difference between the two groups could be found. Twenty-four hours after parturition the differences in the composition of colostrum disappeared, and subsequently no significant difference could be detected.

### References

- Csapó, J., Horváth, A. M., Makay, B. (1982a): Holstein-fríz, magyartarka és magyartarka  $\times$  holstein-fríz ( $F_1$ ) tehenek főcsteje és átmeneti teje szárazanyag-, nyersfehérje-, savófehérje-kazein- és nem fehérje nitrogén tartalmának összehasonlítása (Comparison of the dry matter-, crude protein-, whey protein-, casein- and non-protein nitrogen contents of colostrum and post-colostral milk from Holstein-Frisian, Hungarian red spotted and Holstein-Frisian  $\times$  Hungarian red spotted ( $F_1$ ) cows). *Magyar Állatorvosok Lapja*, **37**, 411–414.
- Csapó, J., Terlaky-Balla, É., Csapó, Zs., Makay, B. (1982b): Holstein-fríz, magyartarka és magyartarka  $\times$  holstein-fríz ( $F_1$ ) tehenek főcstejének, átmeneti tejének és tejfehérjének aminosavösszetétele, valamint aminosavösszetételének változása az ellés után (Amino acid composition of colostrum postcolostral milk and milk protein and their changes in Holstein-Frisian, Hungarian red spotted and in Hungarian red spotted  $\times$  Holstein-Frisian ( $F_1$ ) cows after calving). *Magyar Állatorvosok Lapja*, **37**, 415–419.
- Csapó, J., Csapó, Zs. (1984a): A hungarofríz alapon végzett jersey és holstein-fríz criss-cross keresztezés hatása a kolosztrum és a tej összetételére (Effect of the criss-cross method used for the Hungarofries, Jersey and Holstein-Frisian cattle on the composition of colostrum and normal milk). *Szaktanácsok*, **1**, 32–37.
- Csapó, J., Csapó, Zs. (1984b): A kecsketej fehérjetartalma, fehérjeösszetétele és makro- és mikroelem tartalma (Protein content, protein composition, macro- and microelement of the goat's milk). *Tejipar*, **33** (3), 69–73.
- Csapó, J., Csapó, Zs., Lengyel, A. (1986a): A juh kolosztrumának és tejének összetétele (Composition of ewe's milk and colostrum). *Tejipar*, **35**, (1), 11–24.
- Csapó, J., Csapó, Zs. (1986b): Optimization of hydrolysis and determination of amino acid content in food and feed products. *Acta Alimentaria*, **15**, 3–21.
- Csapó, J., Csapó, Zs., Németh, K. (1987): A kecske kolosztrumának és tejének összetétele (Composition of goat's milk and colostrum). *Tejipar*, **36** (2) 35–45.
- Mancini, G., Carbonara, A., Heremans, J. F. (1965): Immunochemical quantitation of antigens by single radial immuno-diffusion. *Immunochemistry*, **2**, 235–254.
- Moore, S., Stein, W. H. (1951): Chromatography of amino acids on sulfonated polystyrene resins. *J. Biol. Chem.*, **192**, 663–681.
- Morup, K., Olesen, E. S. (1976): New method for prediction of protein value from essential amino acid pattern. *Nutrition Reports International*, **13**, 355–365.

## *Lectures*

# SOIL PRODUCTIVITY AND SOME PROBLEMS OF INTERNATIONAL COLLABORATION AND EDUCATION\*

I. SZABOLCS

RESEARCH INSTITUTE OF SOIL SCIENCE AND AGRICULTURAL  
CHEMISTRY OF THE HUNGARIAN ACADEMY OF SCIENCES, BUDAPEST

(Received 5 May 1988)

The soils of the world constitute the basic resource for food production with respect to both the quantity and the quality of nutrients which directly or through the food chain supply the demand of all living organisms. Nothing can replace soil in this function although it also plays a decisive role as a basic component of the environment which is not only the cradle and cemetery of all living substances but also a buffer which keeps the physical, chemical and biological status of the biosphere in balance. In addition to all this soil ensures the decomposition and often detoxication of huge amounts of compounds produced by agriculture as well as by industry or the communal sphere. It also plays a significant role in the purification of water and provides a foothold for plants, animals and human beings. Still, from the agricultural point of view, the main function of the soil is and remains to be the primary source of nutrition for everything alive, first of all plants.

More than 90% of the biomass of the world, both living and dead, is in the soil. This figure represents a similar proportion of bioenergy.

It follows that all branches of agriculture are, directly or indirectly, related to the soil and their functions, methods and achievements should be adjusted to its productivity. The total area of all the continents are 131 million km<sup>2</sup> but only a little more than 10% of this territory is cultivated.

The distribution of cultivated land on the different continents is uneven. In Asia, for example, nearly 20% of all lands are under cultivation, in Europe this figure exceeds a third, while in Africa it is only a little above 5%. These ratios encourage contemplating the future prospects of solving the food problem in Africa in spite of the fact that the correlation between land use and food production is not as simple as comparing two figures.

\* Lecture held at the C.I.T.A. Agricultural World Congress 4-7 April 1988, Athens, Greece



It is rather difficult to make predictions or even to characterize with figures the potential productivity of the soils of the world but as a general estimate we can state that the soils of the world are capable of supplying its increasing population with adequate amounts of food and raw materials for the foreseeable future.

In spite of the high potential productivity of the soil cover of the globe, there are disquieting signs showing that the present utilization of soils is far from being in harmony with sustainable agriculture. The diminishing of arable land, caused by two kinds of factors, is a world-wide phenomenon.

(1) One of these factors is conversion to other purposes; and the other is the

(2) reduction of arable land by adverse processes.

(1) The conversion of agricultural and silvicultural areas to other purposes is an ever increasing process due to urban and industrial development as well as transportation and the drastic disturbance of land by strip mining, water logging, road and dam construction, etc. In the U. S. more than 2 million hectares of rural land are converted annually to urban and transportation uses and water bodies, and more than 1.5 million ha of fertile soil have been destroyed by mining in the last 40 years. It is predicted that in a few years another 50,000 ha will be ruined just by surface mining. In the GFR nearly 3% of all agricultural land has been lost in the same way in the last decade. Similar tendencies exist in many developing countries where in spite of food shortage often the most fertile soils are subjected to building, roads, canals, airports, etc. Unfortunately putting rural land to other uses is a practically irreversible process leading to the permanent loss of fertile soils.

Governments should urgently limit or arrest the reduction of agricultural land because it is a destruction of renewable resources. However, instead of preventing and arresting the reduction of fertile areas, the same governments often spend large amounts of money on soil reclamation in the same regions which is reminiscent of Sisyphus's task.

(2) Among the adverse processes which greatly reduce the productivity of land we have to mention:

(a) Soil erosion which threatens more than a third of the fertile soils of the world and takes away every year incredible amounts of humus, plant nutrients and other valuable materials.

(b) Secondary salinization, alkalization and water logging of irrigated areas which affect more than half of all irrigated soils and result in the abandonment from production world wide of a territory at least twice as large as the whole of Greece yearly.

(c) The contamination of soils by harmful compounds including acid precipitation, air-borne or water-carried heavy metals, residues of pesticides, radioactive fission by-products, sludge, inorganic toxic materials and residues.



(d) Mechanical and agricultural destruction of soils by heavy machinery.

The facts and considerations outlined above prove that the knowledge and application of soil science are necessary in all branches of agriculture. At present the collaboration between several organizations planning and executing agricultural projects is far from sufficient with the representations of up-to-date soil science. I only want to mention two examples.

(1) It has been referred to that a great part of irrigated lands is exposed to secondary salinization and alkalization. Proper methods of survey and prediction of these processes are available. Decision makers however rarely show interest in them before planning and constructing irrigation systems. Only when the irrigated land has been salinized do they contact soil scientists; which is similar to seeing a doctor only for autopsy.

(2) Another example of insufficient knowledge and collaboration between soil scientists and those who are responsible for production and planning is the failure or lack of protection against erosion during land consolidation, setting up cropping patterns, road building, etc. In most places the reduction or even arrest of erosion would be possible by the application of available preventive methods.

Evidently a better collaboration between the various branches of science and production as well as the organizations concerned must be initiated from all sides and I should like to comment briefly on the biggest world organization of soil scientists, namely on the International Society of Soil Science.

The International Society of Soil Science (ISSS) has eight thousand members from more than a hundred countries and it practically unites all the professional people specializing in this subject all over the world. The ISSS holds a world congress every 4 years and between two congresses it organizes nearly a hundred professional meetings in different parts of the world. In the organization of inter-congress meetings a dominant role is played by the national soil science societies as well as the permanent commissions of the ISSS, namely the Commissions of Soil physics, Soil Chemistry, Soil Biology, Soil Fertility, Soil Classification, and Geography, Soil Technology and Soil Mineralogy.

In addition to the seven commissions, subcommissions and working groups of the society are responsible for raising up-to-date practical subjects and organizing programmes. The ISSS is a member or consultative member of several professional organizations of the UN family and other international scientific bodies (ICSU, etc.).

Science and technology develop rapidly in our age, it is not easy to keep pace with that development. That also applies to soil science and many interrelated questions between it and other branches of agricultural and environmental sciences. This is the reason why education in agriculture should be adjusted to the recent aims and achievements of the sciences includ-



ing soil science. The education of soil science is far from being universal not only in the different countries of the world but sometimes even in one country. Basically the teaching of soil science at university level is conducted mainly in two ways.

(1) In agricultural faculties of universities of agriculture and agricultural high schools.

(2) Faculties of earth sciences or faculties of geography or of geology at universities.

It is impossible in this short paper to characterize the different regions and countries in respect of the above scheme and it is also impossible to interpret and enlist the strengths and weaknesses of either of the two types of education. One aspect however should be mentioned in this place, namely that in the agricultural sphere the education of soil science should be more intensively attached to the achievements of modern basic and environmental sciences than it is at present. It is also necessary to develop the collaboration between the different countries in respect of agricultural education to reach a better understanding of local problems, achievements and failures. There is an increasing demand for the exchange of experiences between the different countries and areas in the field of agriculture as well as for making convertible the terminology, methods and practical systems. Such an international approach and getting familiar with the views of others could help a lot. From a global approach and the adaptation of successful methods developed in different countries both science and production would benefit. This is why education is a crucial problem not only in gaining the knowledge but also in its dissemination.

I hope that the International Confederation of Technical Agricultural Engineers can contribute to the development of up-to-date methods of collaboration and education and the Athens Agricultural World Congress will be a milestone on the road to their realization.

## Reviews

### THE IMPORTANCE OF PLANT BREEDING IN THE RESULTS OF CROP PRODUCTION WITH SPECIAL REGARD TO LUCERNE

I. BÓCSA

RESEARCH INSTITUTE OF THE GÖDÖLLŐ UNIVERSITY OF AGRICULTURAL SCIENCES,  
KOMPOLT, HUNGARY

(Received 14 October 1987; accepted 23 November 1988)

The author, relying mostly on Hungarian literary data, describes the importance of plant breeding in increasing the national yield averages of wheat, maize and sugar-beet crops. Of fodder crops, he chooses lucerne as an example to show that, in a very low rate of increase in the national yield averages, breeding has played a slight role, which is, however, a world phenomenon. He gives full details of the latest objectives of lucerne breeding, including such special questions as the attempts to decrease the proportion of antinutritive (saponin) substances, and to increase the seed yielding capacity, which are much more promising than selection for productivity.

**Keywords:** erucic acid, genetic advance, glucosinolate, *Festuca Lolium*, lucerne, breeding, progress of selection, rape saponin, seed yielding capacity, self-compatibility sugarbeet, wheat

#### Introduction

There is no longer any doubt that plant breeding is an autonomous science. Naturally, when we emphasize its autonomous character — which otherwise is proven by the existence of two departments at our university — it is superfluous to mention its perfect integration with crop production, since the practice of crop production now depends upon the final product of breeding: the variety, furthermore, this becomes increasingly important, a fundamental and indispensable element — so to say the means — of crop production. It is so much so that we encounter increasing difficulties when trying to separate the effect of the variety — i.e. plant breeding — from the agro-technical effects on the increase of yield averages.

The methods of calculating the element of varietal importance in the total growth of average yield are very different. The latest French estimates (Deeprez, 1982) have reckoned with about 50 kg/ha of winter wheat and 30 kg/ha of winter barley for the last 20 years. Between 1960 and 1980 the white sugar yield increased similarly by 50 kg/ha/year in France due to breeding, which indicates a share of some 50% in the increase of average yield with all



three crops. In England the average yield increased by more than 1900 kg of winter wheat and 1200 kg of barley in the last thirty years, from which 1300 kg of wheat and 650 kg of barley (68% and 50%, respectively) are attributed to the varietal effect (cit. Desprez, 1982).

In Hungary similar calculations were first made by Sváb and Szabó (1973) for wheat. They processed 19 years of data on small plot experiments carried out by the Institute Agricultural Variety Testing between 1954 and 1972, using the cumulative yield estimation methods elaborated by Sváb. According to the results 48% of the growth of average yield was credited to plant breeding and 52% per cent to the agrotechnical progress.

Kapás (1978) using linear calculations in his work, indicated that, of the 114 kg/ha annual growth of average yield obtained for winter wheat in 1960–1972, 36% depended upon the variety factor and 64% upon agrotechnics; the corresponding values for maize were 44% and 56%, respectively, in 1963–1972, although one might think that the change of varieties at that time from open pollinated to inbred hybrids ought to have produced a higher ratio than that in favour of the varieties. The failure was due primarily to the fact that the hybrids could show their essentially higher potential productivity only in the state farms, some 20% of the sowing area, while in the co-operative farms, which at that time struggled with everyday difficulties and basic technological problems, even the semi-intensive conditions were not available for the production potential to manifest itself. So the inbred hybrid maize can really be said to have preceded its era.

In recent times such analyses were made by Balla et al. (1985), Balla (1987) and Szabó, Sváb and Baráth (1987) exclusively for winter wheat. Balla et al., and Balla without specifying the methods of calculation, state that in the last 25 years the annual average growth of yield average was 135 and 114 kg/ha, of which 46% and 40–42% were due to the variety. Szabó, Sváb and Baráth (1987) on the other hand, similarly by means of cumulative yield estimation credited only 34% to breeding while attributing 66% to cultural practices out of the 2.4 t/ha and 141 kg/year growth of yield over 17 years. This demonstrates a close conformity to the estimates of Kapás (1978). Finally Szabó (in: Barabás 1987) considers the effect of variety by itself to be only some 25% of the factors determining the yield. It is obvious that the fairly significant differences in estimating the effect of breeding may arise from variations in method, examination period and variety. The terms “effect of breeding” and “progress of selection” are deliberately used instead of the usual term “genetic advance”, which is employed in quantitative or population genetics to denote a component of the genetic variance, that is the additive genetic value, or half of the breeding value (Sváb, 1978). It is not wise therefore to use “genetic advance” instead of “progress of selection”, because with overemphasizing the role of genetics in the trend of the yield



averages, the achievements of plant breeding would be minimized. While genetics is undoubtedly the basic science of plant breeding, plant breeding is an autonomous applied science and as such can be well differentiated from genetics. It is thus more expedient in the future to speak and write of progress of selection or progress of breeding, or occasionally of varietal factor, instead of genetic advance.

It is remarkable that, while in Western Europe the share of variety in the growth of sugar-beet yield is approximately 50%, in Hungary — according to Kapás' estimates — the effect of the variety is only 11% on the root, and 24% on the sugar yield. However, it must be considered that the monogerm triploid varieties, which had a complex effect of on both the varietal and agrotechnical factors, and on their interactions, were not widespread at that time. It is proper to mention here that the triploid monogerm variety Beta M/102 was first registered in Europe in 1968, but the sugar industry — for reasons unknown — did not cultivate it for years; and even in 1975 it was grown on a mere 16% of the sowing area. Today monogerm triploids are widely cultivated, but an analysis of the share of the varieties has not been made for a decade (Csapody and Magassy 1967, Magassy 1969). It must be noted here that the term monogerm is used commonly in the most natural way, and nobody considers the monogerm character as being one of the most outstanding achievements of plant breeding in the last 20 years, a genetically established purely technological property, the *conditio sine qua non* of sugar-beet production in the developed countries. We may even say that without monogerm seed, sugar-beet production would not exist today. It is a breeding achievement equal to male sterility, which is difficult to assess economically.

In any case sugar-beet production, when examined from the standpoint of breeding, has shown a peculiar trend for the last 20 years. With the introduction of triploid varieties, only the root yield increased by some 10 tons, while sugar content stagnated or even decreased to some extent in response to the triploid hybrids, but primarily due to agrotechnical factors including the N overdoses. Beside an unchanged sugar percentage, the white sugar output still increased essentially, which — according to Magassy (1986) — can be put to the credit of breeding and agrotechnics in equal proportions. This matches the above cited French estimation (Desprez 1982). The growth rate of the white sugar yield is, however, not only a result of the increase of root yield. It also depends on how much the quality of beet juice has improved through breeding; that is, it can be attributed to a decrease in the K-, N- and harmful amino-N contents which reduce the sugar output and inhibit the crystallization. Altogether, the share of breeding in the yield growth of sugar certainly exceeds even 50%.



*What is the situation with the fodder crops and planted grasses ?*

Let us examine the situation with the field fodder crops in general, including the species of planted grasses. It is generally true even today that in comparison of the cereals and industrial crops to fodder crops, only moderate if any varietal demands are raised. These demands only appear on a species level, as is the case with the perennial grasses. It happens that, in the absence of sufficient knowledge, the practice cannot formulate the demands, and breeding has to satisfy this function.

With grasses this situation is to some extent understandable. Since grass cultivation in Hungary is on a low level, the planted grasses consist at least of 4, but mostly of 6–8 grass species and a mixture of 2–3 leguminous species, where these are in such complicated interaction and constant competition with each other that assessing the effect of variety would be futile. This phenomenon is connected with the fact that single-species planted grasses — like those in Western Europe — hardly exist in Hungary, and the effect of breeding becomes indistinct in the mixtures. In Western Europe the breeders have achieved considerable successes with tetraploidy in *Lolium* species, and with many other species, primarily tall fescue. In the former crop the quantity of yield is concerned; with the latter, the field of palatability and intake. Further a new conception was introduced: selection for digestibility. In mostly single-species or single-variety grasses the effect of breeding is easier to determine, though studies of this nature are unknown. It is a fact that, in the case of tall fescue, the intake of which by grazing was very low, strain- and clone selections recently carried out through the animals have resulted in a considerable reduction of the silicium content and a consequent improvement in intake, palatability and digestibility (Gillet et al. 1983).

*What is the situation with the lucerne ?*

At last we have arrived at our most important field fodder crop, lucerne, which with its 310,000 ha area still maintains its third place after wheat and maize, though for the last 10 years its sowing area has been reduced by some 120,000 ha, and the very best lucerne areas had to be surrendered to the cereal programme. Its yield average stagnated for 10 years, and has not even increased by more than 1 ton/ha since 1965, the year of the first large-scale plantations. This means that the annual growth of yield is a maximum of 50 kg hay value on a 20-year average, and with the 10-year stagnation also taken into consideration the growth — a mere 25 kg/ha/year — is negligible. Lucerne certainly is an exception among the major crops of Hungary, as in its extremely low growth rate of yield, the average breeding had hardly any share.



Lucerne is known to be an autotetraploid cross-pollinating species of complex heredity, which has resisted so far the breeders' attempt to increase its yielding capacity so much that most of the current registered varieties do not essentially differ in productivity from the ecotypes acclimatized by Tessedik 200 years ago (Bócsa 1981). The best example of this is the local variety Nagyszénási, the exact age of which is unknown, but likely to be at least 100 years old in its present form, a registered variety which under extensive conditions has not been surpassed by any of the more recent varieties. This situation did not change with the discovery of cytoplasmic male sterility in 1960, although it raised great hopes. However, the heterosis effect of TC-hybrids produced by controlled pollination was unfortunately insufficient to make them competitive and economically cultivable. The first Hungarian — and European — hybrid lucerne, the KM-Hybridalfa proved short-lived and could not become commercially widespread, so in 1985 it was cancelled in the variety register.

The fact that breeding has not contributed to the yielding capacity of lucerne was confirmed by a comparison between earlier and more recent French, Swedish, Bulgarian and Hungarian varieties for productivity, as shown in the Hungarian national trials in 1980. It was found that between 25–30 years old (Europe, Alfa, Dunavka and Nagyszénási) and recent varieties of 4 ecologically different lucerne-growing countries there was no essential difference in yielding capacity. That is, genetic advance in this field cannot be discussed (Bócsa 1981). However, at the same time, this phenomenon is not at all a Hungarian peculiarity since nowhere in Europe have there been achieved notable successes in this regard. While out of the other field crop species several dozens of foreign varieties have been introduced for 25 years, not a single foreign lucerne variety has been registered despite the fact that about 150 foreign varieties of the species were tested during this period. In Hungary, the Hungarian varieties were always relatively, even if not significantly better. That is, the lucerne is a rather soil-bound species in which the interaction of origin and ecological factors plays an extremely important role.

It cannot be said that lucerne breeding in Hungary has been totally ineffective. It certainly is not true if the yielding capacity — a characteristic considered by many as the only important one — is left out of consideration. Breeding has been instrumental in maintaining the earlier yield averages in spite of the 100 thousand ha, i.e. 25%, reduction of the total best production areas in the past 10 years, although scientific analyses of this subject have not been completed.

Lucerne varieties which are resistant or tolerant to *Verticillium* and *Fusarium*, that cause the so-called wilt diseases, have appeared in the meantime. Although unable to increase the national yield average, these characteristics have a part in increasing the perennialness, and prolonging the per-



sistence, which are extremely important features in the case of perennial plants. In the seventies when there was a boom in green meal drying, the frequent early cutting, and the so-called wilt diseases of vascular bundles subsequently attacking the weakened stands, reduced the persistence of lucerne to 2.7 years on a national average; but, under intensive conditions of use, it has practically been reduced to a 2-year crop. This indicates an adaptive, indirect effect of breeding, the share of which in the growth of yield average would be extremely difficult to calculate, a reason why nobody has so far undertaken this task. At present we have the courage to declare that, owing to the resistant varieties and through a more careful utilization, not only the persistence has been prolonged by more than 1 year to 3.8 years on a national average, but also the wilt diseases are in a process of retreat. Since the introduction of *Verticillium* resistant varieties, the same has been reported by Guy, the well-known French lucerne breeder and geneticist (Guy 1985).

Even if we cannot speak of successes in breeding for productivity, a feature depending on a great many genes and complicated still further by tetraploidy, well definable features can be successfully approached. These are the already mentioned resistance (resistance to lodging), seed yielding capacity persistence, tolerance to frequent cutting, and last but not least absence of antinutritive elements, which depend on much fewer factors and are less influenced by the environment. Results worth mentioning have certainly been attained in this field. However, from the techniques applied to increase the yielding capacity such triumphs as experienced in the case of maize or winter wheat in the last 25 and 15 years, respectively, cannot be expected in the foreseeable future.

The selection for tolerance to frequent cutting is a good example of breeding for adaptive features. This objective gained priority in the 1970s, when intensive utilization became a question of primary interest. It was found that almost every population possessed an exploitable genetic variability which, however, remained latent to manifest itself only after 4-5 cuts. By recurrent selection of plants tolerant to frequent cutting, the population's response can be changed in the course of 3-4 cycles, and rapidly regrowing populations yielding more with 1-1.5 additional cuttings, without losing their persistence, can be produced. Another great advantage of these varieties is the higher protein content compared to other varieties, due to the fact that they can be cut in an earlier phenophase when the crude protein content is higher and the crude fibre percentage lower. Thus, the biological antagonism between dry matter yield and protein content existing primarily in the vegetative plant parts can be circumvented, and the otherwise unattainable objective of breeding for protein content indirectly approached without any decrease of persistence in the selected population.



One great discovery of the last decade is that fodder crops in general and lucerne in particular can be successfully bred for absence of the so-called antinutritive components which are noxious for both monogastric animals and humans. The presence of antinutritive substances in lucerne was discovered more than two decades ago, but their reaction mechanism has not yet been established. Only their final effects and consequences are known. These are weight gain- and growth inhibition and reduction of egg production in the case of saponins, as well as inhibition of amino acid absorption in the case of tannins (polyphenols) (Mårtensson 1979). Furthermore, there are growth- and reproduction disorders caused by certain flavonoids and cumestrols through oestradiol in mammals (Montier and Rambourg 1978). Out of these antinutritive factors of known structure but partly unknown biological reaction mechanism, saponins are undoubtedly the most important. These triterpenoid pentacyclic compounds are in fact glycosides of sapogenins. The most important biologically active sapogenin is the medicagenic acid (Cheeke 1971).

A wider use of lucerne in feeding monogastric animals is mainly prevented by the saponin content. If lucerne was to be fed to monogastric animals in lieu of a considerable amount of soybean, first the so-called leaf extraction technique had to be elaborated, which has been known in Hungary as Holló-Koch i.e. Vepex leaf protein producing procedure for some 15 years. This method produces a protein concentrate containing 50% crude protein, —2% cellulose, and other components of extreme value compared to soybeans, such as  $\beta$ -carotene and xanthophyll, the latter a very effective colouring agent used in poultry industry and egg production. The concentrate called by us LFK (APC, alfalfa protein concentrate is equal in value with the soybeans, because, though it contains less methionine, the carotene- and xanthophyll contents absent in soybean compensate for the methionine deficiency. The only great disadvantage of the LFK is its saponin content that makes its use impossible for poultry farming and human nutrition. It is unnecessary to mention how important it would be to the starving third world, not only for its protein content but also as a natural source of vitamins and minerals.

Having read the promising American publications which referred to the high heritability of the saponin content, we started selection from relatively low saponin content varieties of a world collection in 1972. We isolated groups of varieties with relatively low saponin contents, determined the range of variability of the saponin content, the high temperature dependence of the saponin synthesis, year effects, etc. (Buglos et al. 1981). A number of biological determination tests suitable for quick serial analyses (blood haemolysis, *Trichoderma*, *Tenebrio*, *Lebistes*, etc. tests) had to be adapted. With their help we picked out the minus variants over 4 cycles, and in the 4th cycle selected material containing 1/4–1/6 of the amount of saponin found in the standard varieties, without any decrease in yield. This amount of saponin was biologi-



ically inactive as practically proven by the experiments carried out at Gödöllő in 1985–1986, with several hundreds of broiler chickens (Vetési 1985). Thus, for the time being this product with very low saponin content of high biological value might be used in the poultry branch where a third of the imported soybeans could be replaced by LFK. Of course this requires building up the so-called infrastructure. That is, the old lucerna-drying facilities now existing should be completed with machine lines for the extraction of leaf protein. Costs of the investment realising this plan would be high but it solved the replacement of soybeans from a basic material produced at home. We have also started to multiply Szapko, a variety registered in 1987 as the first and only low saponin variety of Europe or perhaps even of the world (Bócsa et al. 1986).

A further — mainly theoretical — task is to produce a zero-saponin material which will not show saponin reaction to biological tests. Moreover, saponin will not be detectable in it even with the latest techniques used in analytics. Thus we should like to discover what role saponin, as a secondary metabolic product, plays in the lucerne plant, and to prevent possible physiological and agronomical defects. Namely, some are afraid that the low saponin lucerne will be more susceptible to diseases and pests, as well as losing its frost resistance and winterhardiness. It should be mentioned otherwise, that in the 3-year national tests the low saponin variety Szapko has always matched the standard varieties as regards productivity.

### Conclusions

With the varieties free from antinutritive components, further difficulties arise in assessing the role of breeding and its share in commercial production, since in this case a new variety opens up new vistas by making a non-conventional utilization possible. It cannot be compared to the current varieties either for yield or for nutritional quality as it is not an additional feature, but it is a valuable deficiency that we encounter here which qualifies the variety for a certain use. Likewise it cannot be said that only saponin deficient lucerne varieties should be grown in the future, since ruminants do not need such varieties because the microflora of the rumen totally decomposes the saponin and in any case ruminants would not anyway be fed with the very expensive LFK.

Out of the species grown on larger areas, winter rape, for which elimination by breeding of certain not totally antinutritive components like erucic acid and glucosinolate, plays an important role. The harmfulness of these components to the human organism is widely discussed; their carcinogenic effect has not been unambiguously proven in experiments with rats, yet, to



meet the demands of the always subjective market, erucic acid-free or -deficient varieties are required. Also, the chemical industry needs varieties with very high erucic acid content (Eöri 1986). The breeder is therefore forced to follow two diametrically opposed but parallel objects of breeding. It is established that the so-called glucosinolate (mustard oil by its former name), when present in meals and cakes, is harmful. In response to certain enzymes this compound decomposes to such further compounds which have a deleterious effect on the thyroid gland and the reproductive phase, and for this reason rape-meal and rape-cake cannot be unlimitedly fed. Without these compounds, and — naturally — in the case of a further decrease in fibre content, the rape-meal would be nearly equal in feeding value to the soya meal, which would be of very great importance. In foreign countries there exist varieties with 1/10 of the amount of glucosinolate contained in the standard varieties (Obidzinski 1985).

Finally we wish to speak of breeding for increasing the seed yielding capacity, in lucerne. It is a rather new objective of breeding, not even mentioned in Hanson's monograph on lucerne published in 1972, although results in this field are known to have been attained earlier in America. The fact that Hanson makes no mention of it suggests that even in the USA there is no uniform standpoint on the question. In Hungary seed yielding capacity was not until recently recognized as an important varietal feature, the general view being that lucerne is not grown for its seed. It took a long time to prove that the seed yielding capacity is an important feature in a plant species the seed yield of which hardly exceeds 100 g/ha on a 15 year average, and the annual scatter may be as much as 70 kg. Today seed production in its proper place is acknowledged as an important varietal character, though it has not yet been included in the official descriptions of varieties. It is obvious that out of two varieties with the same potential fodder production the one that gives an essential larger seed yield will be agronomically more valuable. American authors were the ones who pointed out that no negative correlation existed between fodder yield and seed production, as shown by the example of the internationally well-known variety Europe with its excellent green matter production and high seed yielding capacity.

Selection for seed yielding capacity offers good results, since variation in this character among 10 varieties chosen at random from any gene bank may even be 100%, while the difference in fodder production between the best and the poorest variety is a maximum of 10%, as seen in the 1980–1982 *Eucarpia trials* carried out with 14 varieties in 8 countries (Rod et al. 1986).

The difference in seed yield between the best and the poorest variety was 66% on a 3-year average. This indicates that variability is very high even on the level of variety without any selection. Among the Hungarian varieties e.g. Verko yields 30% more seed, compared to the average of all Hungarian



varieties. According to calculations made at our Institute (Pummer 1986) with seed yield taken twice a 4-year period, a variety like that exceeds in agronomical value the other varieties by some 10% in 4 years. That is to say, its production ensures a much higher income.

Two methods are suggested for the work of breeding. One of them is to maintain a recurrent selection for number of seeds per pod; that is, to increase the main parameter of fertility, the fertilization percentage, while the rate of pods as a secondary factor naturally also kept in consideration. In the case of the mentioned variety Europe this course is followed, while the vigour and the maximum potential of fodder production are invariably maintained. The other means to success leads through a radical change in the flowering- and fertilization biology of the lucerne plant. It is to the self-pollinated sour-cherry varieties that such a lucerne variety can be best compared, with its inclination to self-tripping, high self compatibility, self fertility and at the same time tolerance to inbreeding depression.

Finally, it should be emphasized that the varieties as products of plant breeding on the one hand, and plant breeding itself as a scientific-pare artistic activity on the other must be regularly assessed for their economic utility. It would be highly desirable not to restrict this assessment to a few plant species, mainly to wheat and maize, but rather to extend it to the major field and horticultural crops. Any further support for and development of plant breeding can only be expected if its usefulness, indispensability and scientific position will be continually proven.

### References

- Balla, L. (1987): Az akadémiai búzanesemítés eredményei (Results of academic wheat breeding). *Magyar Tudomány* XXXII. 4, 301–309.
- Balla, L., Szunics, L., Szilágyi, G. (1985): *Genetikai haladás a búzanesemítésben* (In: Bajai, I. és Koltai, Á. szerk: Búzatermesztési kísérletek 1970–1980). (Genetic advance in wheat breeding (In: Bajai I. and Koltai, Á. eds. Wheat growing experiments 1970–1980)). Akadémiai Kiadó, Budapest.
- Bócsa, I. (1981): *Amélioration du rendement en matière sèche chez la luzerne*. Compte rendu de la Conférence Eucarpia Group Medicago sativa. Kompolt 1–3. July, 1980.
- Bócsa, I., Sárosi, J., Fehér, F., Majkó, Z., Vetési, M. (1986): Szaponinszegény lucernafajta nemesítésének eredményei egygyomrú állatok fehérjeszükségletének helyettesítésére (Results of breeding lucerne for low saponin content to substitute the protein requirements of monogastric animals). *Növénytermelés*, 35, 287–292.
- Buglos, J., Bócsa, I., Manninger, K., Manninger, S. (1981): *Saponin content and its relationship to variety, temperature and field resistance to Fusarium and Verticillium fungi in alfalfa*. Report of the twenty-seventh alfalfa improvement conference, Madison, Wisc. 80–81.
- Cheeke, P. R. (1971): Nutritional and physiological implications of saponins. A review. *Can. J. Anim. Sci.*, 51, 621–632.
- Csapody, M., Magassy, L. (1967): Ergebnisse und Probleme der Züchtung pollensteriler monokarper Zuckerrüben in Ungarn. *Tag. Ber. DT. Akad. Landwirtschaft.-Wiss.* Berlin, 89, 255–272.
- Desprez, M. (1982): L'amélioration des plantes, ses résultats, sa situation actuelle, ses perspectives d'avenir. *Fertilisants et Agriculture*, 83, 21–29.
- Eöri, T. (1986): *A repce termesztése* (Rape cultivation). Mezőgazdasági Kiadó, Budapest.

- Giller, M., Noël, Cl., Jadas-Hécart, J. (1983): La cafétéria d'auges, méthode d'étude de l'appétabilité. *Agronomie* 3, 817–822.
- Guy, P. (1985): Verbal information.
- Hanson, C. H. (1972): *Alfalfa science and technology*. Amer. Soc. Agron. Madison.
- Kapás, S. (1978): *A fajtaváltás hatékonysága* (Efficiency of the change of variety). Akadémiai Kiadó, Budapest.
- Magassy, L. (1969): A fajtafenntartás és javítás módszereinek fejlődése, továbbá lehetőségei a répanemesítésben (Development of the methods of variety, maintenance- and improvement, and their possibilities in sugar beet breeding). *Agrártud. Közl.* 26, 381–391.
- Magassy, L. (1986): Szóbeli közlés (Verbal information).
- Martensson, P. (1979): *Studies on tannin, saponin and tripsininhibitors in lucerna*. Eucarpia Fodder Crops. Section Meeting, Radzikow, 294–301.
- Monties, B., Rambourg, I. C. (1978): Présence de flavonoïdes sensu stricto (flavones et coumestanes) dans des préparations de protéines extraites de luzerne (*Medicago sativa* var. Europe). *Ann. Technol. Agric.* 27, 629–654.
- Obidzinski, W. (1985): Dwuzerowy rzepak-bialkowa szansa na przyszłosc. *Przegląd Hodowlany* 53, 14–17.
- Pummer, L. (1986): Szóbeli közlés (Verbal information).
- Rod, J., Pelikán, I., Bócsa, I. (1986): *International variety trial with lucerne grown for seed*. Eucarpia Medicago Sativa Group. INRA, Paris.
- Sváb, J. (1971): *A populáció genetika alapjai* (Fundamentals of population genetics). Mezőgazdasági Kiadó, Budapest.
- Sváb, J., Szabó, M. (1973): A búzatermés növekedésének vizsgálata terméslelemzéssel, 1954–1972. években (Examination of the increase in wheat yield by yield analyses in 1954–1972). *Növénytermelés*, 22, 289–300.
- Szabó, M., Sváb, J., Baráth, C. (1987): A búzatermés 1969–1985 közötti növekedésének szétválasztása környezeti és nemesítési komponensekre terméslelemenként (Distribution by yield component of the growth factors of wheat yield in 1969–1985 between environment and breeding). *Növénytermelés*, 36, 73–82.
- Szabó, M. (1987): *Fajtakérdés, fajtarotáció, fajtavédelem*. (In: Barabás, Z. szerk.: A búza-termesztés kézikönyve) (Question of variety, rotation of variety, protection of variety. (In: Barabás, Z. ed: Handbook for wheat growing)). Mezőgazdasági Kiadó, Budapest.
- Vetési, M. (1986): *Szaponinmentes lucernalevél fehérjekoncentrátum etetése broiler csirkékkel* (Saponin-poor lucerne leaf protein concentrate fed to broiler chickens). Gödöllő.





## RECENT PROBLEMS OF AMELIORATION OF SALINE AND ALKALI SOILS

I. SZABOLCS

RESEARCH INSTITUTE FOR SOIL SCIENCE AND AGRICULTURAL CHEMISTRY  
OF THE HUNGARIAN ACADEMY OF SCIENCES, BUDAPEST, HUNGARY

(Received 8 February 1988)

About 10% of the surface of continents is covered by different types of salt affected soils. The utilization and reclamation of these soils is a recent problem in many countries. Saline and alkali soils are the most frequent types of salt affected soils and their amelioration demands the application of up to date and reasonable methods according to natural and economical conditions of the given area.

Some of principal problems of reclamation methods are reviewed in this paper.

### Introduction

About 10% of the total surface of our globe is covered with different kinds of salt affected soils which have two common features:

1. the effect of electrolytes on the soil forming processes, and
2. low fertility.

Salt affected soils are common not only in the vast areas of deserts and semi-deserts but also in fertile river basins, sea shores and other regions where the salinity or alkalinity of the soil constitutes a barrier to production. This is why the amelioration and utilization of salt affected soils have first rate importance in many places of the world, both in developing and developed countries, in dry or humid areas, in agri-, sylvi- or horticulture and under different conditions of production.

The existing more than 10 million square kilometres of salt affected land includes vast territories of so-called secondary salt affected soils, which have become salinized due to application of improper methods of irrigation from ancient times to our days.

The amelioration and improvement of salt affected soils have a long history in the various countries, mainly in those which have been confronted with the problem for a long time. Due to the different environmental and ecological conditions, the different necessities and possibilities, innumerable methods of reclamation have been developed and applied. By now it has become difficult in many cases to interpret and evaluate them, and to decide upon the method to be applied in one or in the other place for the best utilization of salt affected soils. This is the reason why an up-to-date review is needed to characterize the problem and some possibilities of its solution.



Among the different types of salt affected soils the most common and extended are the saline and alkali soils which jointly amount to more than four-fifths of all existing salt affected soils in our globe.

Amelioration methods have been elaborated and projects realized mainly for improving the mentioned two types.

### A brief characterization of saline and alkali soils

It is generally accepted that water soluble salts, particularly the sodium salts, are responsible for the low fertility of salt-affected soils. Soils whose content of salt (or their ions) interferes with the growth of the majority of crops are termed saline or alkali soils.

Two main groups of such soils have been distinguished:

(1) Soils affected by neutral sodium salts (mainly sodium chloride and sodium sulphate)

(2) Soils affected by sodium salts capable of alkaline hydrolysis (mainly  $\text{NaHCO}_3$ ,  $\text{Na}_2\text{CO}_3$  and  $\text{Na}_2\text{SiO}_3$ ).

Soils belonging to the first group have mainly been named saline, and those of the second group alkali soils. These two main types differ not only in their chemical character, but also in their geographical and geochemical distribution, as well as in their physical, chemical, physico-chemical and biological properties. The methods used for their reclamation and agricultural utilization are also different.

Although it is evident that in nature the various sodium salts do not occur absolutely separately in soils, in most cases either the neutral sodium salts or the ones capable of alkaline hydrolysis exercise a dominating influence on the soil-forming processes and soil properties.

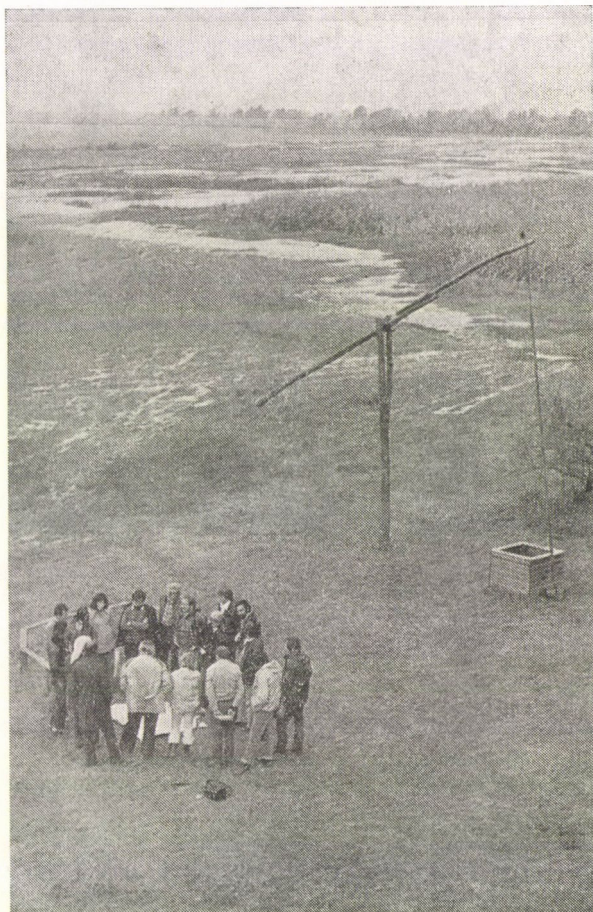
On the various continents, under the very wide range of environmental conditions, the general levels of the salinity or alkalinity of parent materials and ground waters may sharply differ. The salinity or alkalinity tolerance of local crops varies widely too. The potential salinity or alkalinity of an area depends, to a considerable extent, on the cropping systems used there. It is more than obvious that in all these respects only very vague limit values can be given on a world-wide scale. Therefore it is necessary that — while keeping the basic principle in mind — a certain flexibility be employed in the definition of salinity and/or alkalinity limit values characterizing the salt-affected soils of a given territory, that is, the local conditions should also be taken into consideration.



### *Saline soils*

On most continents saline soils are the dominant type of salt-affected soils. In Europe, however, the situation is different; here the alkali type prevails.

In saline soils the dominating salts, mainly  $\text{NaCl}$  and  $\text{Na}_2\text{SO}_4$ , reach sometimes one or more per cent of the soil material, particularly in surface layers, but usually we speak of saline soils where the above mentioned value is above 0.5%. In many countries the electrical conductivity (E. C.) value is used for the measuring of salinity, and if this exceeds 4–6, saline phase or saline soil is diagnosed. Without going into particulars about the determination methods of salinity, this soil type is to be briefly characterized.



*Fig. 1. Saline soil*



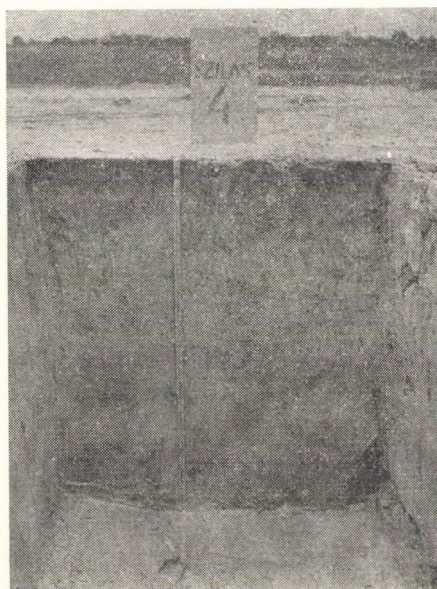


Fig. 2. Profile of a saline soil

The maximum of salt accumulation may be found at different depths in the soil profiles but very often it occurs in the top layer or near the surface. The saline soils have developed in the most arid regions. The few exceptions to this rule are caused by the salinity of local ground water or soil forming substrata.

Figure 1 demonstrates the surface of a saline soil. Seasonal changes effected by the climate and, particularly, by irrigation and drainage, may occur in the salt contents of saline soils, as well as in the distribution of salts in the different layers of the soil profile.

The basic morphological feature of saline soils is the lack of a structural B horizon. Although several morphological systems use the letter "B", in those cases it never signifies a horizon distinguishable from the A horizon by its well-developed structural formation. Consequently, the profiles of saline soils are rather monotonous, from the surface down to the parent material. In few cases, when saline soils have formed under bog conditions, the top layers are humous, but usually, when they have developed under arid conditions, these soils are very poor in humous substances and their humus content is lower than 1%. A low plant nutrient content (mainly N and  $P_2O_5$ ) is also characteristic of most saline soils.

The high salinity determines practically all physical and chemical properties of saline soils, consequently, when these properties are evaluated,



the salt content of such soils and its influence should first of all be taken into consideration.

Many classification systems employ the term "chloride and/or sulphate solonchak" for saline soils.

Figure 2 shows the profile of a saline soil.

The soil surface is covered with a thick layer of water-soluble salts and along the profile there are abrupt boundaries of different morphological horizons.

### *Alkali soils*

In alkali soils the presence of Na salts capable of alkaline hydrolysis determines the soil properties. Due to their effect either the high alkalinity of the soil solution hinders plant growth, or the alkalinity renders the physical soil properties disadvantageous for the water supply of plants. Evidently often both of these processes exert their harmful influence, though in alkali soils without structural B horizon the latter dominates.

As a rule, in alkali soils without structural B horizon a considerably high concentration of sodium salts capable of alkaline hydrolysis — mainly sodium carbonate — may be found. Of all these salts which commonly occur in soils, sodium carbonate has the most harmful effect on both soils and plants. Thus this subclass represents salt-affected soils which have very disadvantageous properties for agriculture. Their fertility, if any, is very low. Not only the alkalinity, but in most cases, also the salinity of these soils is rather high. This is why in many classification systems they are denominated "alkali-saline" or "saline-alkali" soils. Although sometimes neutral sodium salts may prevail among the water-soluble substances in these soils, neverthe-



**Fig. 3.** A spot of alkali soil in the Hungarian Plain



less in most cases the dominant role is played by sodium carbonate owing to its high alkalinity.

The alkali soils which have structural B horizon in their profile are commonly named solonetz. The B horizon has a well-developed structure, mainly columnar. It can be easily distinguished from the horizon above (A horizon), which is less compact and whose structure is less developed. This



Fig. 4. Solonetz profile

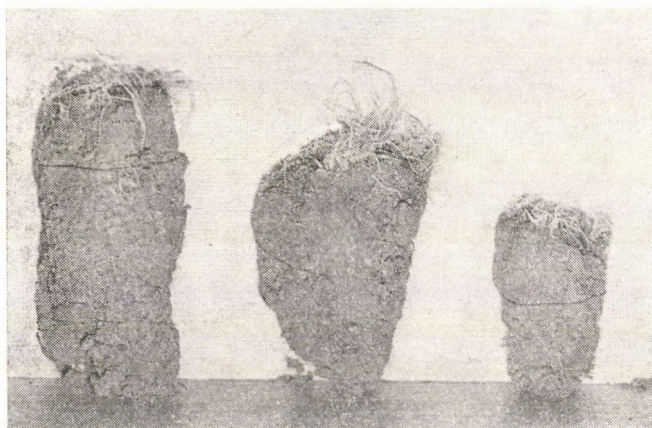


Fig. 5. Columns of solonetz soil

B horizon determines the genetic type of these soils, their main physical, chemical, physico-chemical and biological properties, as well as their fertility together with the possibilities of their agricultural utilization.

Figure 4 represents a solonetz profile which is a well-developed columnar structure.

A structural B horizon always markedly differs from the A horizon not only in morphology, colour and structure, but also in its physical, chemical, physico-chemical and biological properties.

In Figure 5 columns of solonetz soil are demonstrated.

### Main aspects of reclaiming saline and alkali soils

The reclamation method of a saline or alkali soil must be decided first of all after considering the climatic conditions.

Saline and alkali soils in different places and under different conditions have different characteristics parallel with their common features. These characteristics must be taken into consideration when elaborating the methods for their reclamation.

The main characteristic properties of saline and alkali soils are described in Table I.

From the detailed characteristics given in Table I the following can be inferred:

(1) In saline soils, the high content of sodium salts often shifts the pH value toward the alkaline reaction. However, this value as a rule does not

Table 1  
*The characteristics of saline and alkali soils*

Characteristics	Saline soils	Alkali soils
pH	8.3	8.3 somewhere along the profile, or ESP > 15 in horizon B
Chemistry of soil solution	Dominated by SO <sub>4</sub> and Cl anions	Dominated mainly by HCO <sub>3</sub> or CO <sub>3</sub> anions or both
Effect of electrolytes on soil particles	Flocculation	Dispersion
Main adverse (or toxic) effects on plants	High osmotic pressure of soil solution	Alkalinity of soil solution
Geographical distribution	Associated mainly with semi-arid areas	Associated mainly with semi-arid and semi-humid areas
First aim of reclamation	Removal of excess electrolytes through leaching	Lowering or neutralizing the high pH through chemical amelioration



surpass 8.3, except in some particular cases. On the other hand, in many saline soils, especially when a considerable amount of  $\text{CaSO}_4$  and  $\text{CaCl}_2$  are present, the pH value may be lower than the neutral level.

In alkali soils with structural B horizon (solonetz soils) carbonates are frequently missing, and as a consequence, the pH is below 8.3, particularly in the surface layer or near the surface of the soil. In the B horizon of these soils the ESP value is high and, as a rule, exceeds the value of 15. However, in some places, it is somewhat less than 15 whereas in other cases, the value surpasses this percentage, mainly in solonetz soils.

(2) In saline soils, the sulphate and chloride anions dominate whereas in alkali soils bicarbonate and carbonate anions prevail. It must be noted that in extremely arid regions nitrates may play a dominant role among the anions of the electrolyte.

In general, alkali soils without a structural horizon always contain free alkali carbonates while alkali soils with a structural B horizon do not have free carbonates in all cases. Thus we may distinguish carbonate-free and sodacontaining solonetz soils.

(3) The main effects of electrolytes on particles, especially on soil colloids in saline soils is coagulation. Consequently in these soils the water-transmission properties, such as the infiltration rate and hydraulic conductivity are more favourable than in alkali soils, in which the alkali-hydrolysing electrolytes exercise a dispersive effect. These differences underline the different reactions of alkali and saline soils to drainage and irrigation.

(4) In saline soils, the high osmotic pressure of the soil solution, as a result of high electrolyte content, constitutes the main harmful effect on plants (physiological effect). On the other hand, in alkali soils it is the poor physical and water-transmission properties of the soil which most frequently impede the growth of plants (soil physical effect). The toxic effect of different ions ( $\text{Fe}^{2+}$ ,  $\text{Mn}^{2+}$ ,  $\text{S}^-$ , etc.) may be felt in both cases, particularly under waterlogged conditions. It should be noted that sometimes the high electrolyte content also exercises a toxic effect. For example, the relative toxicity of seven salts occurring in salt-affected soils is roughly as follows:

MgSO <sub>4</sub>	CaCl <sub>2</sub>	Na <sub>2</sub> SO <sub>4</sub>	NaCl	NaHCO <sub>3</sub>	MgCl <sub>2</sub>	Na <sub>2</sub> CO <sub>3</sub>						
1	:	1	:	2	:	3	:	3	:	4	:	10

(5) In arid regions, mainly saline soils can be encountered whereas in a moderate climate, the alkali soils dominate among the salt-affected soils. This is a general rule, but evidently, saline soils may be found in some moderate areas too, if the soil-forming factors (e.g. the high electrolyte contents of the sediments and underground waters) lead to the development of soil salinity. Likewise, the alkalinity of soils may occur exceptionally in desert



and semi-desert regions (e.g. as a consequence of high carbonate contents of sediments and waters).

(6) Different approaches should be adopted in the utilization and improvement of saline and alkali soils. The first step of reclamation should be to control the dominant adverse properties. In the agricultural utilization of saline soils, high electrolyte content is the main obstacle to increasing soil fertility and, therefore, the establishment of drainage and leaching are the preconditions of effective amelioration.

### Reclamation of saline soils

In the course of reclaiming saline soils, excess salinity has to be removed, or at least diminished. For this purpose leaching and drainage are two basic methods. The only practical way, according to our present knowledge, to remove excess soluble salts from the soil is by washing them out. Evidently provisions must be made for disposing of the water in order to prevent re-salinization.

Drainage draws the water off, thereby improving saline soils. A proper drainage system for saline soils must be designed to desalinize not only the top soil layer but also the upper subsoil and water-bearing horizons. It will regulate both the water and salt balances of the soil and subsoil.

Various types of drainage are used: vertical, deep-horizontal, shallow horizontal, etc. Vertical drainage has given good results in certain soil conditions, particularly when the deep horizons are highly permeable. Shallow horizontal drainage is still widely used in spite of the fact that it is not always successful.

The main methods for the reclamation of saline soils are very similar in all countries. There are 3 preconditions of effective amelioration:

- (1) To keep the water below the critical depth by drainage.
- (2) To reduce the soluble salt content to 0.4—0.5% in top soils and 2–3 gs/litre in the groundwater. This process can be controlled by irrigation, leaching and drainage.
- (3) To avoid re-accumulation of toxic salts in the root horizon, if necessary, by repeated leaching.

In the USA leaching and drainage are planned as far as possible to keep the upper 1 1/2 metres of the soil free of excess salts. Detailed methods are elaborated for the effective reclamation of saline and alkali soils. The so-called Handbook No. 60, edited by the US Salinity Laboratory, Riverside, California, comprises the recent knowledge and methods in this field.

Similar methods have been elaborated in the Soviet Union where the effective leaching of solonchaks under conditions prevailing in Central Asia



and in the Caucasus takes place in late autumn or in winter when the soil moisture is low and the water table is deep.

The leaching process should begin on lower laying areas and work up gradually onto higher ground.

The application of gypsum or other chemicals (sulphur, calcium chloride, calcium nitrate, etc.) is inevitable in the reclamation of alkali soils, particularly where they lack the structural B horizon.

Vast territories of saline soils have been improved during the last decades. Unfortunately no exact data are available on the sizes of these soils. On the other side due to improper methods of irrigation, similarly large territories have been salinized during this time. The conclusion can be drawn that the extent of saline land has not diminished in the arid regions. Still, it is a fact that many efforts and attempts were devoted to the elaboration of proper methods for land reclamation and they brought both scientific and economic results.

### Reclamation of alkali soils

Alkali soils often occur in areas where the precipitation and other conditions are very favourable for rain-fed agriculture and only soil alkalinity hinders good production.

From the aboves it can be inferred that the reclamation of alkali soils must be more diverse and specific than that of the saline soils.

It is a general principle that before the reclamation of an alkali soil it is to be determined whether it has or has not a structural B horizon.

In many classification systems alkali soils are named solonchak solonetz, solonetz, or solod soils. The dominating group among these types is the solonetz. That is why the terms "alkali soil" and "solonetz soil" will be used in the following as synonyms.

The solonetz soils in a dry region should be reclaimed either under irrigated or non-irrigated conditions. The degree of aridity and the required yield determined whether the reclamation can be successful without irrigation.

In moderately or non-arid areas the groundwater conditions play an important role not only in the formation of alkali soils but also in their utilization.

In the technical literature widely differing data may be found concerning the utilization and reclamation of solonetz soils in the various countries. These differences are caused partly by the widely varying local conditions (climate, parent material, level of agricultural technology, etc.). The different salt profiles and salt dynamics — with particular regard to the depth and the quality of the groundwater — exercise a decisive influence not only on the genetics of solonetz and solod soils but also on the possible methods of their



amelioration and utilization. In this respect these soils may be subdivided into three main groups:

(1) In the case of solonetz and solod soils where the soil profile and the top layers are capillarily linked with salty groundwater and the horizons ( $A_1$ ,  $A_2$ ,  $B_1$ ,  $B_2$ ) contain about 0.2% or more water soluble salts in the surface layer, and 0.5% at a depth of 40–50 cm, the leaching out of salts and drainage are unavoidable. In this case the reclamation of solonetz and solod soils may be similar to that of solonchak (saline) soils.

As regards leaching, it may be carried out either by applying irrigation water of good quality and by providing good drainage or — under more humid conditions, when the annual precipitation is enough to leach out the salts — by lowering the water table below the critical level. Chemical amendments should also often be applied parallel with the above mentioned measures or afterwards, especially in the case of heavy textured soils, in order to replace the adsorbed  $Na^+$  in the colloidal fraction by calcium. In certain cases solonchak-solonetz soils may also be reclaimed this way.

As regards the genetics of solonetz and solod soils, mainly the meadow solonetz and meadow solod soils belong to this group.

(2) If the profile of a solonetz or solod soil is only temporarily linked with groundwater, and the salt content of the A,  $A_1$  and B horizon is lower than that of the soils belonging to the first group, drainage is not always necessary. In these cases the application of chemical amendments (gypsum and/or others) as well as deep-ploughing and loosening of the subsoils may be useful. If the  $B_2$  and C horizons the quantity of water soluble salts is not high and a considerable amount of gypsum is present, in the course of deep-ploughing it can be utilized as reclamation material. Depending on the local conditions, amelioration may be carried out either with or without irrigation. Under irrigated conditions, however, the providing of good drainage is more important. Soils belonging mainly to this group are often called meadow solonetz and solod soils turning into steppe formation.

(3) If the profile is not linked with the ground water, the salt content in the upper layers of the profile should be the most important consideration when the suitable amelioration method is selected. In these cases when the present, natural soil formation processes assure, to a higher or lesser degree, the leaching out of water soluble salts, drainage must be satisfactory. Frequently these soils are only moderately solonized and/or solodized. Their exchangeable  $Na^+$  content is less than 10–15% of the cation exchange capacity, and it may be found in a comparatively deeper layer, at more than 15–20 cm below the surface.

In the case of these soils, it is possible to employ non-expensive and simple reclamation methods with good results because the basic aim of reclamation is to facilitate natural leaching out processes. The climatic conditions,



the possibility of irrigation, etc. are also decisive factors when the proper methods are chosen to remove the salts and to improve the physical soil properties.

Naturally in reclamation of both saline and alkali soils, the local environmental conditions and economic possibilities have decisive role on selection the proper and rational method. Not only the soil properties, but also the availability and cost of reclamation technics and amendment should be taken into consideration when selecting the design and technology of amelioration for saline and/or alkali soils.

### Selected references

- Alekseevsky, E. E. (1971): *Irrigation and drainage of the World*. Kolos. Moscow. R.
- Antipov-Karataev, I. N. (1953): *Reclamation of Solonetz Soils in the USSR*. Acad. Sci. USSR, Moscow.
- Kovda, V. A. (1946-1947): *Origin and Regime of Saline Soils*. Acad. Sci. USSR, Moscow.
- Kovda, V. A., C. van den Berg, Hagan, R. M. (1967): *International Source Book. Irrigation and Drainage of Arid Lands. Salinity and Alkalinity*. FAO-UNESCO, Paris.
- Kovda, V. A., Szabolcs, I. (1979): Modelling of Soil Salinization and Alkalization. *Agrokémia és Talajtan*, **28**, Suppl.
- Loveday, J. (1985): Soil Salinity and Sodicity in Australia. *Agrokémia és Talajtan*, **34**, 179-184.
- Richards, L. (1954): *Diagnosis and improvement of saline and alkali soils*. US. Dept. Agric. Handbook No. 60.
- Salinity problems in the arid zones*. Proc. Teheran Symp. Arid Zone Res., **14**, 1-395. UNESCO, Paris, 1961.
- \*Sigmond, E. (1927): *Hungarian Alkali Soils and Methods of their Reclamation*. Univ. California, Berkeley.
- Szabolcs, I. (1961): *A vízrendezések és öntözések hatása a tiszántúli talajképződési folyamatokra (The Effect of Drainage and Irrigation on the Soil Formation Processes in the Region Beyond the Tisza in the Great Hungarian Plain)*. Akadémiai Kiadó, Budapest.
- Szabolcs, I. (1964): Salt Affected Soils in Hungary. *Agrokémia és Talajtan*, **14**, Suppl. 275-306.
- Szabolcs, I. (1969): Reclamation methods developed for solonetz and solod soils with a view to their formation process. *Acta Agron. Acad. Hung.* **18** (3-4), 339-346.
- Szabolcs, I. (1974): *Salt-affected soils in Europe*. Martinus Nijhoff, The Hague and Research Institute for Soil Science and Agrocultural Chemistry of the Hungarian Academy of Sciences, Budapest.
- Szabolcs, I. (1979): *Review on research of salt-affected soils*. UNESCO.
- Szabolcs, I. (1987): The global problems of salt-affected soils. *Acta Agronomica Hungarica*, **36** (1-2), 159-172.
- Szabolcs, I., Darab, K. (1982): Irrigation Water Quality and Problems of Soil Salinity. *Acta Agron. Sci. Hung.*, **31**, 173-194.
- USSR/UNEP Project 1982. *Studies on the Impact of Agricultural Management on the Environment*. Summary of Research Results. GKNT, Moscow.
- Worthington, E. B. (1977): *Arid land irrigation*. Pergamon Press, ICSU.
- Zavaleta, G. G. (1965): The Nature of Saline and Alkaline Soils of the Peruvian Coastal Zone. *Agrokémia és Talajtan*, **14**, Suppl. 415-425.



## Book reviews

---

*Advances in Soil Science* Volume 6. Edited by B. A. Stewart Springer-Verlag New York, Berlin, Heidelberg, London, Paris, Tokyo, 1987.

This volume of the series deals with highly important questions of soil science, such as soil chemistry and the problem of nutrient replacement.

The *first chapter*, "Potassium Dynamics in Soils", was written by Donald L. Sparks. The author starts with a description of the characteristics of potassium and its forms in the soil, showing the interactions of the different forms of potassium in figures and the methods of determination in a table (water soluble-, exchangeable-, non-exchangeable-, mineral- and total K content). He examines the dynamic balance between the different K-forms, and by a detailed analysis of the soil colloids proves that the quantity and quality of the latter is the parameter that has the greatest influence on the processes. Methods of measuring the dynamics of K, and as a valuable part of the chapter models and equations describing K dynamics are given. Comparing the dynamic method with the kinetic one, the author points out that the kinetic approach may have many advantages, first of all that in this case information is obtained on the speed and mechanism of the motion of potassium.

The *second chapter*, "Models to assess the susceptibility of soils to excessive compaction", written by S. C. Gupta and R. R. Almaras discusses a question highly important for the practice of agriculture. Owing to the intensive cultivation, big tractors and heavy

machines, the compactness of soils causes serious world-wide problems. After giving exact definitions the authors present the laboratory analyses and describe the apparatus suitable to measure compaction too. They characterize the laboratory model, then evaluate the role of the different factors which act on compaction, and through which excessive compaction causes problems in the soil-plant system. (A decrease in the quantity of pores filled with air may be an obstacle to gas exchange, at a certain pressure the structure of soils will be damaged, the resistance of the soil hinders the growth of roots.)

Laboratory measuring and models are insufficient in this case since models suitable for use under field conditions are also required. The authors describe two models, the system based on pressure distribution in detail, with its application in practice.

The occurrence in soil of *Li*, *Na*, *K*, *Rb*, *Ca* and their role and quantity are the subjects of the *third chapter*: "Sources, amounts and forms of alkali elements in soils". A. D. Scott and S. J. Smith, the authors, summarize the chemical properties, geochemistry, and mineral forms of the elements belonging to the group of alkali metals. The chapter gives more particulars of elements which usually occur in very small quantities in soils and are thus of little interest from the standpoint of soil science. The distribution of alkali metals is given by geographical situation, particle size and soil profile. The forms of elements in soils are specified and discussed as soluble-, exchangeable-, non-exchangeable- and fixed forms.

The *fourth chapter*: "The diagnosis and



recommendation integrated system (*DRIS*)" is a work by J. J. Walworth and M. E. Sumner.

One of the most important tasks of modern applied agrochemistry is to provide quick and reliable advice on fertilization. A promising system in this field is the *DRIS*, which is based on leaf analysis data. Since the concentrations of the various nutrients change in the course of the vegetation period, the system was elaborated for the application of proper nutrient ratios. The authors offer examples of the *DRIS* norms and of the method for preparing diagnoses. They also show how to determine the so-called *DRIS* index, then on the basis of the nutrient indices provide the determination of the minimum factor. The *DRIS* norms are tested by fertilization experiments under glass house- and field conditions. A comparison of the *DRIS* method with others reveals that this system is much more sensitive. The *DRIS* can be extended to a wider scope and applied not only to the nutrient ratios but even to soil analysis data.

Joe Kubota, Ross M. Welch and Darell Van Campen wrote the *fifth chapter*: "Headed soil-related nutritional problem areas for grazing animals". They make a general survey of the nutritional problems of plants and grazing animals in regard to microelements. The micro- or trace elements are divided into four groups, according to the need of plants and animals for them; then the deficiency symptoms, and occasional toxic effects characterizing the elements of each group are described. The nutrition supplying problems of grazing animals are shown by geographical location, now only in respect to four elements; namely, *Co*, *Mo*, *Cu* and *Se*. Tabulated summarization, detailed treatment and chart representation show that problems are caused by the deficiency of *Co* at 0.04–0.07  $\mu\text{g}/\text{mg}$  toxicity of *Mo* above 10–20  $\mu\text{g}/\text{mg}$  deficiency of *Se* below 0.05  $\mu\text{g}/\text{mg}$  toxicity of *Se* above 4–5  $\mu\text{g}/\text{mg}$  concentration in plants.

All this is evaluated in the USA, with the soilforming rock, the characteristics of soil and the vegetation taken into consideration.

The topicality of the subjects and the

high level of the treatment guarantee that a valuable book comes again to the reader's hand.

MONIKA TAKÁCS

ERICK KERRIDGE: *Trade and Banking in early Modern England*, Manchester University Press, 1988, Great Britain 185 p.

Eric Kerridge, eminent author of numerous studies on agricultural and industrial economics remained again true to his scope of subject and to himself alike. Trade and Banking when translated into Hungarian can be better approached by the expression: circulation of commodities and money. Nevertheless, the author's view of the evolution of market — first of all of the market of a big city — is very instructive. He gives a peculiar definition of the essence of market. Accordingly, it is a matter of a voluntary spontaneous exchange of various people's properties, in terms of mutual advantages, under price conditions mutually agreed upon, with the purpose of satisfying each other's actual and supposed demands. This way a special market develops for each group of product, but these markets jointly merge in the universal system of market. Somewhat unlike the other countries of Europe in England the complex markets of big cities began to come into existence in the 16th century, first of all in London. London took up and dispatched considerable volumes of commodity in the Middle Age already. This role was made possible partly by its infrastructure, partly by the means and methods of payment. As to the infrastructure, the conditions of roads had to be improved above all. Namely, at the beginning the commodities were mostly transported by pack-horses, except in summer. It helped much that from the 1600's the communication roads were kept in repair by workers paid by the owners of big estates, from then on the two- and later the four-wheeled vehicles were able to convey much larger quantities. The load of a cart sometimes drawn by 8–10 horses may have even been 1.5–2.5 tons. Another important



step was the introduction of postal service mainly during the reign of Henry VIII.

At the beginning the sluggish circulation of money was an inhibitory factor. It was roughly after 1560 that the use of bill began to spread. The substitution of cash gave a great stimulus to trade. By the turn of the 16th and 17th century bills at long sight also became common means of payment. At the same time the conditions of payment became stricter. In the first half of the 16th century the ratio of cash to bill or bill of debt was 1 : 9 on an average, that is, the bill showed a marked preponderance over the cash. True though, that it was an average of rather extreme values, since the 1 : 26 ratio of merchants was opposed by the distrustful low ratio of 1 : 2 of the land-owner nobility.

To support his statements the author lists 296 sources of literature, and uses more than 300 selected works as additional material to his work. Orientation among his authentic data is facilitated by a detailed subject index.

Kerridge's style is particularly remarkable: the high scientific level is combined with an extraordinary clarity of expression.

#### I. DIMÉNY

T. C. HARRINGTON and F. W. COBB, Jr. (Eds): *Leptographium Root Diseases on Conifers*. APS Press, St. Paul, Minnesota 1988, 1-149.

The book is based, in part, on papers presented at the Reno meetings (August, 1985, Reno, Nevada, USA).

One chapter was included additionally, discussing species of *Leptographium* in Western Canada. As it is stressed in the preface, each paper was accordingly updated to include all the new information until 1987. Increased recognition of root diseases on conifers caused by *Leptographium* spp. (teleomorph: *Ceratocystis* in sensu lato) and growing confusion in the taxonomy of anamorphs and teleomorph of these fungi were the main reasons to present the current status on these diseases.

Attention at the Reno meeting was focused on the main pathogens: *Leptographium wagneri* and *L. procerum* causing black-stain root disease and procerum root disease.

The book collects the most important, current information on world-wide distribution of these diseases and *Leptographium* species, as well as on biology, taxonomy, pathogenicity, hosts, insect associations.

A valuable paper by Harrington T. C. provides a full list of all the recognized taxa (34 species) of *Leptographium*.

We are convinced that this special book will be an excellent basic source, especially for forest pathologists, mycologists engaged in the study of forest tree diseases caused by *Ceratocystis* in sensu lato fungi and their anamorphs including *Leptographium* species.

L. VAJNA

*Biological and Cultural Tests for Control of Plant Diseases* Vol. 3. (Ed.: J. R. Hartman) Amer. Phytopathol. Society Press, St. Paul, Minnesota, USA, 1988. 100 p. ISSN 0887-2236 (88).

This is the third volume of an APS (American Phytopathological Society) series which was developed to disseminate information on the performance of cultivars, biological control agents, and cultural practices for plant disease control, and to improve biological and cultural testing and reporting.

Starting with a special report on Measuring Disease Intensity by R. D. Berger, this volume comprises eighty-three contributions placed into nine sections as follows: Fruits and Nuts, Vegetables, Corn and Sorghum, Soybeans, Small Grains, Field Crops, Turfgrass, Ornamentals and Trees, and Citrus and Tropical. In the first, a single report on the effect of ammonium phosphate and bacterial antagonists on apple replant disease is given. The next section Vegetables is the largest one covering 27 reports that, in the majority, deal with resistance testing of bean, broccoli, cabbage, celery, cucumber, muskmelon, potato, squash, sweet corn and tomato against var-



ious viral, bacterial or fungal pathogens. In the next two sections, cultivar response tests are described of corn and sorghum to different pathogens and of soybeans to cyst nematodes, respectively. Except two reports on pathosystems with rice, section Small Grains includes 16 tests of wheat for resistance reaction to powdery mildew, *Puccinia* leaf rust, *Helminthosporium* root rot, *Septoria* leaf and glume blotch, *Pyrenophora* tan spot, take-all disease, *Fusarium* scab, *Fusarium* head blight, *Rhizoctonia* root rot, wheat streak mosaic, wheat spindle streak mosaic, and wheat soilborne mosaic. Testing methods shown in section Field Crops are mostly related to cultivar response, either alone or combined with other determinants such as various crop management programmes. Two additional contributions in this section provide with biocontrol tests: a rapid screening of bacteria for biocontrol of *Phytophthora* damping-off on lucerne, and effects of *Trichoderma* seed treatment on *Rhizoctonia* damping-off and root rot in sugarbeet. A short section deals with resistance tests of turfgrasses and this is followed by section Ornamentals and Trees where evaluation of cultivars, biocontrol agents and fertilization are equally reported for crops as China aster, crabapple, peperomia and snapdragon. The last section describes pineapple gummosis by *Fusarium moniliforme* var. *subglutinans* in relation to planting date.

The volume is completed with a general index and a list of materials under trial. Plant pathologists, nematologists, plant breeders and all those engaged in plant disease control will find this publication particularly useful in their work.

F. VIRÁNYI

BÁN, I.: *Biomathematics and its applications in plant cultivation*. Akadémiai Kiadó, Budapest, 1988.

This book is a welcome addition to the growing literature of the application of mathematics to biological problems.

Biomathematics may be looked upon as a

borderline between biology and mathematics. It includes analyses carried out by mathematical methods and taking biological uses of procedures in applied mathematics. Biomathematics is a useful tool for discovering regularities, as described in this book.

The book consists of two main parts.

In the *first part* some problems are introduced of biomathematical theory, such as:

- basic biomathematics;
- mathematical approximation of probabilistic processes in the biology on the basis of a set of observed data;
- biological defect of the regression analysis based on the last squares principle;
- role of cognizability limit in detecting the changes of biological state;
- comparison of sets characterizing biological states on the basis of magnitude-relation;
- a multivariate examination method with relative coordinates;
- evaluation of the effect of a treatment using modulating with respect to control.

In the *second part* examples of the mathematical solution of problems in plant protection are presented. For example the establishment of the optimum sampling ratio of the values of certain plant protection characteristics is shown here (Section 2.1.).

When examining plant protection efficiency it is an essential problem whether there is a relationship between the average yield of the grown crops and the plant protection expenditure, and if so what is its regularity. By the known methods of regression analysis it has been proven that, considering the "plant protection expenditure in the average percentage" characteristic as independent variable, and the "average yield in the average percentage" characteristic as dependent variable, there is a significant relationship between the two characteristics (Section 2.2.1).

The other essential problem in plant protection efficiency investigations is whether there is a correlation between the number of plant protection measures and fruit export ratio and, if so, what is the regularity of this relationship. Having assumed the % value



of the fruit export ratio as an independent variable, and the number of control measures as a dependent variable, by the known methods of regression analysis a significant relationship is demonstrated between the two characteristics (Section 2.2.2).

Examinations of plant protection efficiency also cover the examination of farm management indices. In the function of plant protection expenditure, as an example, the farm management indices of raspberry were examined, namely the production cost and grass production value as well as net income dependent variables. In the course of the examination the method "approximating by polynomial a set following different functions types by intervals" was applied. (Section 2.2.3).

The main condition of rational plant protection is the knowledge of the ecological requirement of the injuries and infections of living organisms. The mathematical analysis offers considerable aid in deciding which factors are related with the incidence and other characteristics of the pests and pathogens, as well as the regularities of this relationship. As an example, the relationship between ecological conditions and individual density of the cucuid meal beetle, the rice weevil and of the "confused" flour beetle was examined as a function of the temperature and moisture content of cereals (Section 2.3.1).

Examinations of the ecological requirement of injurious and infectious living organisms are highly important regarding the forecast of the probable rate of injury or infection. As an illustration, the ecological requirement of the late blight of potatoes and the possibility of the mathematical forecasting of the infection were examined (Section 2.3.2).

Assaying the plant protecting agents is one of the major duties in the control of pests and pathogens. By the mathematical analysis of the collected data the protective dosage has to be established. For this the mathematical model describing the regularity of the effect of the plant protection must be established. As an example, first the dose of sodium-pentachlorophenol providing protec-

tion against *Coniophora cerebella* on Scotch fir was determined (Section 2.4.3).

The intention of the book is to acquaint the reader with the problem to be solved and so assist him to reach the solution, (by manual calculation or by using the possibilities of his microcomputers). The draft formulation of practical examples and the control computertechnical solutions is illustrated in this book by detailed illustrations.

This book is recommended to students of forestry, pharmacology, agrotechnics, medicine and natural history in general and experts working in these fields who like new approaches and who like to cogitate over practical problems.

MAGDOLNA PERCZEL

PEARSON, R. C. and A. C. GOHEEN (Eds.), *Compendium of Grape Diseases*. APS Press, Minnesota, USA 1988. 93 text-pages (with 30 figures and 3 tables) and 24 pages with 188 colour illustration, softcover. ISBN-O-89054-088-8.

This is the latest issue of the "Disease Compendium Series" published by the American Phytopathological Society. This Compendium of grape diseases was compiled by 48 authors, all well-known specialists on the subject.

In the *Introduction* authors describe pathogens of grape, species of *Vitis* used in breeding program and as rootstocks, structure and growth stages of grapevine and historical significance in grape production. In subsequent sections diseases are arranged according to causal agent. Diseases caused by biotic agents such fungi, bacteria, mycoplasma, viruses and nematodes are presented in *Part I*. Diseases caused by fungi are subdivided according to the major part of the plant affected (fruit and foliar-, minor foliar-, wood and root diseases).

*Part II* covers some forms of mite, thrips, leafhoppers, treehoppers and *Phylloxera* injury that could be mistaken for disease.

*Part III* deals with disorders caused by abiotic factors, such as nutrient deficiencies,



environmental stresses (drought, excess water, heat, hail, spring frost etc.) air pollution and chemical toxicities.

*Part IV* discusses how cultural practices influence disease.

*Part V* deals with selection, registration and certification of grapevines and regulation of international trade in grapevines for propagation. A list of equivalent disease names in France, German, Italian and Spanish is given in the Appendix. A very useful glossary of viticultural and plant pathological terms will aid all readers. The exact index provides an easy handling of this compendium.

The figures and colour plates unequivocally present the symptoms, the pathogens and in some cases the life cycle of the pathogens.

The compendium was written for those with limited training in viticulture and pathology. For readers who want a deeper knowledge of the subject the selected references at the end of each description are helpful. This Compendium of grape disease is an indispensable guide to a better diagnosis of grape disorders. It can be recommended to growers, crop advisers, extension specialists, plant pathologists and students.

I. TÓBIÁS

*Information on maize.* (Információnnüj Bjületeny po kukuruze. — Koordinacionnüj Centr. SZEZ po probleme KOC-2. Naucno-issledovatel'szkij Inst. Sz/H. Van Marton-vásár. No-6 1987). (Received: 14 April, 1988)

The Agricultural Research Institute of the Hungarian Academy of Sciences, Marton-vásár, which co-ordinates the maize research of the COMECON continuously published the research results attained of the participating countries. Vol. 6. of 1987 contains 24 papers on a total of 391 pages. Eight papers (*Chapter 1*) deal with the effect of various agrotechnical methods on the yield of maize, 15 papers describe recent results in hybrid seed-grain production (*Chapter 2*). In *Chapter 3* (1 paper) the members of the co-ordinator institute characterize the ecological experiments

of the COMECON countries in 1981-1986, evaluating the results by a new biometric analysis. In *Chapter 1* the authors discuss the results of agrotechnical studies, the effect of plant density, the production potential and water demand of maizes of different maturity groups grown on chernozem (Hungary-DATE), the effect of cold test and grain quality on germination and on the yield of hybrid maizes (Hungary-Szeged), the role of the nutrient status of maize (Hungary-Nádudvar), the experiences of the KSZE production system (Hungary-Szeksárd). Other researchers show the correlation of plant number and dry matter production in silage maize (Poland-Szmolica), discuss production technology questions, such as production conditions, variety, forecrop, soil preparation, sowing, plant protection, fertilization (Yugoslavia-Zagreb). One of the main features of the research activity is its many-sidedness.

In the introduction the editors point out that variety is the cheapest and most efficient factor of the agricultural production; further, that high yielding varieties of good quality and agronomical value will also be much sought after in the future. The present varieties require specific agrotechnics. In the first chapter of the volume the optimum sowing time, fertilization, water supply, crop rotation, effect of herbicides under experimental and in production conditions (experiences of production systems and farms) are dealt with. Hungarian authors point out the role of crop rotation, seed-crop monoculture, to the correlation between soil preparation and yield, and give percentage data on the share of the major factors (fertilizer, variety, plant number, soil cultivation, plant tending) in yield increase (Györffy B.—Berzsenyi Z.). Others call attention to the special water reactions of hybrids as well as to the correlation of water and nutrient supply (Nagy, J.).

The availability of nutrients greatly depends on the hybrids. The effect of irrigation depends also upon the water economy of the soil (Ruzsányi L.). On the part of production systems the condition of the soil (Co-operative Farm, Nádudvar), and the role of machine



technics — including the new biotechnics, and computer technics — are emphasized (KSZE, Szekszárd). Hungarian researches discuss the role of seed-grain quality, injured seed, soft- and hard seed in germination (Szél, E. et al.). Foreign authors deal with maize cultivation in their respective countries (Pucarics, A. et al., Yugoslavia, Zagreb). Polish researchers (Krulikowskij, Z. et al.) give the optimum plant number for silage maize production in norther socialist countries, with a view to increased dry matter production.

All researchers support their statements with a large number of data (tables, figures).

The authors deal with many questions of the seed-grain quality of hybrid maize in further 15 papers of *Chapter 2* some important subjects are: the effect of plant density for inbred lines on the quantity and quality of grains (Bulgaria-Knezsa), relation between the biological value and specific weight of seed-grain (Hungary-Mv), the effect of plant density on the shape and size of the maize seed-corn (Hungary-Szeged), the effect of stand density and harvesting time on the quality of seed-corn (Hungary-Mv). In this chapter the authors deal with technological tasks of production (Truksa Jn, Czechoslovakia-Trnava), the effect of stand density on seed-grain quality under different agroecological conditions (Yugoslavia-Zagreb), the state of water loss of lines and hybrids during maturing (Yugoslavia-Zemun-Pole, Belgrade) with the use of inbred lines and sister-lines in hybrid seed-corn production Yugoslavia-Zemun-Pole, Belgrade), injuries to the pericarpium in processing (Yugoslavia-Zemun-Pole, Belgrade), the present situation and prospects of seed production in Yugoslavia (Zagreb), with new conceptions in studying the physiology of the hybrid seed-grain (Yugoslavia-Zemun-Pole, Belgrade). Further, they discuss the effect of the fungicides TMTD and Kaptan on the initial development of maize lines (Yugoslavia-Zemun-Pole, Belgrade), the interaction between seed quality and yield in hybrid maize (Yugoslavia-Novi Sad), the effect of nitrogen and irrigation on the yield of inbred lines (Yugoslavia-Zemun-Pole, Belgrade). Grain processing- and cultivation expe-

riences with ZP-hybrids (Zemun-Pole) are evaluated on the basis of experiments carried out at the Zemun-Pole institute (Yugoslavia-Zemun-Pole, Belgrade). The reaction of the 2 line to increase plant number in seed production depends up its genotype though the reaction should be studied on the level of hybrid as well. The genotypic yield components change to a lesser extent (Preszolszka, P.).

Hungarian authors point out that the plat number influences the yield and the parameters of grain processing alike. The harvesting time affects not only the quality of seed but also the expected heterozis effect (Záborszky, S.-Szundy, T.). Gabrics, I., (Yugoslavia) says that the grain yield and its quality is influenced by the fertility of soil, its physical and chemical properties, the quantity and distribution of rain as well as the variety and its critical water demand, the plant number depending on the maturity group. These are fundamental agrobiological conditions. Others deal with the water loss of inbred lines at the period of maturing (Kerecki, B.—Koics, L.—Ratkovics, S.) with possibilities offered by the sister-lines (Pavlov, M.), with the shape- and size dependent injury of grains (Popovics, P.) and draw attention to the importance of handling chemical reagents carefully so as to preserve the quality of the seed. It makes a difference where and to what extent the seed becomes injured. Physiological questions and preservation of the viability of the seed during storage are also mentioned Radenovics, C. et al. Others point out the influence of fungicides (TMTD, Kaptan) and the different responses of inbred maize lines to them (Sztankovics, M.—Pavlov, M.). Some authors discuss the relationship between nitrogen and irrigation (Vasic, G. et al.), or the relation of the quantity and quality of the hybrid seed (Simin, V.,—Bogdanovics, B.). An account is given of maize hybrid production experiences in Yugoslavia (Pucaric, A.—Gotlin, J. and Vidojkovics, M.—Popovics, R.—Csernicsek, L.). Researchers from the northern socialist countries summarize their experineces of grain-, silage- and CCM utilization.



The researches described in this chapter stress the theoretical bases of hybrid seed production, and the practical questions of this cultivation and processing, i.e. the whole vertical chain of the branch.

In Chapter 3 some Hungarian authors (Martonvásár) report on ecological experiments carried out in the framework of the COMECON KOC<sub>2</sub> project. An important aspect of the evaluation is the difference in climatic conditions between the socialist countries taking part in the experiment, which may greatly influence the average and must therefore be taken into consideration despite the fact that the experiments are carried out according to maturity groups.

The volume gives comprehensive information on current problems of maize cultivation and seed-grain production on the basis of the research results of the socialist countries engaged in maize cultivation, and on the subjects of research in maize cultivation and on the subjects of research dealt with in 1981–1986. Many useful experimental and production data are published in it concerning the relationship between agrotechnics and yield, and their factors, respectively. This new COMECON-KOC<sub>2</sub> publication with its information on the research lines and — experiences is equally useful for those concerned either with the theory or with the practice of the corn specialist.

A. KOVÁCS

WULF DIEPENBROOK and MARGIT FRENTZEN, *Membranlipide (Struktur, Lokalisation und Biosynthese sowie agroökologische Bedeutung und landwirtschaftlichen Kulturpflanzen)*. Paul Parey Scientific Publisher (Berlin, Hamburg, 1988) brought out as the 9th volume of "Advances in Agronomy and Crop Science" an extremely useful book for the interest of many representatives of the profession.

The work of 106 pages is divided in two parts. In the first part the structure, localization and biosynthesis of the membrane lipids are dealt with: in the second, the larger part discusses the importance of the variability of polar lipids.

In the introduction the authors briefly explain the importance of this subject. Changes in the composition of the membrane lipids — as a basic mechanism — make the cultivated plants adaptable to changes in the surrounding temperature, which in the higher plants is a much more complex feature than in those on a lower level of organization. The authors refer to the discovery of Raison (1973) namely, that the high unsaturation degree of the lipid fractions in hardy species is an indicator of "fluidity", an important requirement for the membrane function. According to the results obtained so far there is a correlation between adaptability and the fatty acid composition of membrane lipids, though it is often inconsistent. The changes in the lipid composition are manifold and varying, depending on external influences and on the age of the tissues examined.

The agroecological importance of the question is due to the possibility that the adaptation of agricultural and horticultural crops can be recognized in the membrane lipids.

As a matter of course, the degree of unsaturation of their total amounts in a given tissue is less characteristic than those specific changes which occur in some lipid groups and characterize the structure of certain cell compartments (as e.g. the glycolipids do the chloroplasts). From an agroecological point of view, the authors wish to answer the question of how much the variability of the membrane lipid properties is determined by external factors, as a result of which the conditions of senescence in the different organs are established.

#### *Structure, localization and biosynthesis of membrane lipids*

The length of a recension being limited there is no way to deal — even if in outlines — with the aspects given in the title. We only want to point out that the readers are given thorough information not only on the structure and biosynthesis of the large number of polar lipids but also on their share and distribution in the membranes of various crops. The sketches showing the pathway and



localization of the biosynthesis help the comprehension not only in the case of eukaryotes but also with prokaryotes. The various membranes systems are characteristic with lipid fractions of asymmetrical position: typical and significant differences can be seen, e.g. in the structure of membranes inside and outside the plastids.

*Agroecological importance of polar lipids*

In this larger part of the book the authors deal with the lipids of leaf, root and seed in separate chapters. In the case of leaf lipids the influence of temperature and nitrogen, then the interactions of light, temperature and nitrogen are discussed. Numerous graphs and histograms make the experiments generally carried out with crops (*Zea*, *Hordeum*, *Brassica*) — and exceptionally with trees (*Pinus*) for the sake of comparison — suggestive.

In the chapter on root lipids, brief theoretical data are given concerning the ions entering the symplast from the intercellular space; then, the importance of nitrogen,  $\text{Ca}^{2+}$ -ions and pH is discussed.

Extremely instructive are the result of experiments carried out with the "split-root" technique in hydroponic cultures in order to determine the effect of different ions on the

characteristics (fatty acid composition, degree of unsaturation etc.) of the root.

The seed lipids (in the cotyledons and endosperm) mostly are triacylglycerides characterized by hydrophobia and an extremely low acid content. The seed lipids are considerably affected by the temperature, e.g. low temperature increases their degree of unsaturation. While in the leaf and root the temperature induced changes of lipids can be brought into connection with adaptation, the importance of unsaturation in the seed lipids is unknown. For example, the three most important unsaturated fatty acids (oleic acid, linolic acid and linolenic acid) give different responses to changes in temperature (e.g. in *Brassica* species), which undoubtedly suggests that in the biosynthesis of these fatty acids, physical factors — e.g.  $\text{O}_2$  concentration in the tissues — are exclusively involved. Nevertheless, it is highly probable that the enzyme activation and biosynthesis have genetic conditions as well.

The book is completed by a short English summary. The highly valuable and thoroughgoing book was written with some 600 literary works used as sources. The attractive design and the quality of figures and paper do credit to the editor.

I. SZALAI





## REVIEWERS OF MANUSCRIPTS VOLUME 37 1988

Every scientific contribution in *Acta Agronomica Hungarica* is reviewed by two scientifically qualified persons. The Editorial Board is pleased to publish the following list of reviewers for the manuscripts of the 1988 issues, who by their unselfish contribution have significantly contributed to ensure the scientific standards of *Acta Agronomica Hungarica*.

Balázs, Sándor  
Balla, László  
Bálint, Andor  
Bernáth, Jenő  
Boross, László  
Bócsa, Iván  
Brunner, Tamás  
Bubán, Tamás  
Dános, Béla  
Debreczeni, Katalin  
Dohy, János  
Erdei, Erzsébet  
Filius, István  
Frenyó, Vilmos  
György, Beattie  
Hamar, Norbert  
Hargitai, László  
Herold, István  
Heszky, László  
Hornok, László

Horváth, György  
Jenczer, Gábor  
Kiss, Árpád  
Kiss, Á., Sándor  
Kiss, Violetta  
Kovács, Antal  
Lásztity, Borivoj  
Lelley, János  
Manninger, István  
Máthé, Imre, Jr.  
Nagy, Béla  
Nagy, József  
Nyéki, József  
Obadovics, György  
Pais, István  
Pál, István  
Pásztor, Károly  
Pecznik, János  
Pethő, Menyhért  
Précsényi, István

Sáringer, Gyula  
Simon, József  
Solymosi, Péter  
Surányi, Dezső  
Szabady, Judit  
Szabó, Gy. László  
Szabó, Miklós  
Szalai-Marzsó, László  
Szegi, József  
Szlovák, Sándor  
Tétényi, Péter  
Tomcsányi, Pál  
Tóth, Imre  
Tölgyesi, György  
Tuba, Zoltán  
Varga, Magdolna  
Völgyi, József  
Vörös, József  
Wellich, Péter





mouton de gruyter

Berlin · New York

# europaean review of agricultural economics

**Editor:** Kees Burger  
Economic and Social Institute  
Free University, Amsterdam, The Netherlands

The **European Review of Agricultural Economics** serves as a forum for discussions about the development of theoretical and applied agricultural economics research in Europe and for stimulating ideas regarding the economic problems of agriculture in Europe and other parts of the world.

The **ERA** also promotes discussion on national resource use, protection of the environment, marketing of agricultural products and development of rural areas.

Throughout, the **ERA** strives for balanced coverage of all issues in agricultural economics: production economics, operations research and farm management problems, agricultural policy, including farm incomes and farm structure, regional planning and rural development, supply analysis, factor markets, demand analysis and marketing, international trade and development, statistical and econometric methods, etc. Original articles as well as full or abstracted articles which have already appeared in national publications and/or in other languages are included. Shorter features supplement the main contents and ensure that the most recent information available is covered. These features include research notes, book reviews, comments on previously published articles and news items about European activities in the field of agricultural economics such as meeting and conferences.

The **European Review of Agricultural Economics** is published as one volume for four issues per year (approximately 520 pages).

## Subscription rates for Volume 16 (1989):

Institutions/libraries	DM 198.00
Individuals (prepaid only*)	DM 101.00 (includes postage)
Single issues	DM 54.00

## Prices in US\$ for subscriptions in North America only:

Institutions/libraries	US\$ 110.00
Individuals (prepaid only*)	US\$ 43.00 (includes postage)
Single issues	US\$ 38.00

\* Individual subscriptions are for personal use only and must be prepaid and ordered directly from the publisher. Prepayment may be made by cheque or by credit card: MasterCard (Access), EuroCard, Visa, and American Express (AMEX may not be used in North America). The individual rate is not available in the FRG, Switzerland, or Austria.

Subscriptions, single, or back issues may be ordered through your local bookseller or subscription agent, or directly from MOUTON DE GRUYTER (a division of Walter de Gruyter) at either of the following addresses:

**For North America:**  
Walter de Gruyter Inc.  
200 Saw Mill River Road  
Hawthorne, N.Y. 10532  
USA

**For all other countries:**  
Walter de Gruyter & Co.  
Postfach 11 02 40  
D-1000 Berlin 11  
Federal Republic of Germany





**mouton de gruyter**

Berlin · New York

## **european review of agricultural economics**

**Editor:** Arie Oskam  
Agricultural University  
Wageningen, The Netherlands

The **European Review of Agricultural Economics** serves as a forum for discussions about the development of theoretical and applied agricultural economics research in Europe and for stimulating ideas regarding the economic problems of agriculture in Europe and other parts of the world.

The **ERAE** also promotes discussion on national resource use, protection of the environment, marketing of agricultural products and development of rural areas. Throughout, the **ERAE** strives for balanced coverage of all issues in agricultural economics: production economics, operations research and farm management problems, agricultural policy, including farm incomes and farm structure, regional planning and rural development, supply analysis, factor markets, demand analysis and marketing, international trade and development, statistical and econometric methods, etc. Original articles as well as full or abstracted articles which have already appeared in national publications and/or in other languages are included. Shorter features supplement the main contents and ensure that the most recent information available is covered. These features include research notes, book reviews, comments on previously published articles and news items about European activities in the field of agricultural economics such as meetings and conferences.

The **European Review of Agricultural Economics** is published as one volume of four issues per year (approximately 512 pages).

**Subscription rates for Volume 17 (1990):**

Institutions/libraries	DM 198.00
Individuals (prepaid only*)	DM 116.80 (includes postage)
Single issues	DM 57.00

**Prices in US\$ for subscriptions in North America only:**

Institutions/libraries	US\$ 115.00
Individuals (prepaid only*)	US\$ 46.70 (includes postage)

\* Subscriptions for individuals are for personal use only and must be prepaid and ordered directly from the publisher. Prepayment may be made by cheque or by credit card: MasterCard, (Access), EuroCard, Visa, and American Express (Amex may not be used in North America). Orders placed for institutions will be invoiced at the institutional rate. The individual rate is not available in the FRG, Switzerland, or Austria.

Institutional subscriptions, single, or back issues can be ordered from your local bookseller or subscription agent or directly from MOUTON DE GRUYTER, (a division of Walter de Gruyter) at either of the following addresses:

**For North America:**  
Walter de Gruyter Inc.  
200 Saw Mill River Road  
Hawthorne, N.Y. 10532  
USA

**For all other countries:**  
Walter de Gruyter & Co.  
Postfach 11 02 40  
D-1000 Berlin 11  
Federal Republic of Germany

PRINTED IN HUNGARY

Akadémiai Kiadó és Nyomda Vállalat, Budapest





# AUTHORS' GUIDE FOR MANUSCRIPT PREPARATION

## GENERAL INSTRUCTION

Two copies of the manuscript and two sets of the figures should be submitted to:

Acta Agronomica Editorial Office,  
Ménesi út 44.  
H-1118, Budapest

Manuscripts in English or in Hungarian including Abstract, References, Tables and Legends should be typed double-spaced (25 lines, 50 characters per line including spaces) and supplied with authors' names, page number. Tables should be on separate, numbered pages after the References. Legends for figures, on a separate page, should follow the tables. Standard articles should not exceed seven pages.

## FORMAT

**Title.** The title should reflect the most important aspects of the article, in a preferably concise form of not more than 100 characters and spaces.

**By-line.** The authors' names should be followed by affiliations and addresses. (No inclusion of scientific titles is necessary.)

**Abstracts** are required for all the manuscripts. They should be typed in one paragraph and limited to max. 200 words. Below the abstracts, an alphabetical list of keywords should be given.

**Text.** Major sections after the introductory statements are: *Material and methods*, *Results*, *Discussion*, *References*. Subheadings may be used, though the unnecessary fragmentation of the text should be omitted.

**Style.** After acceptance for publication, manuscripts are reviewed for style, grammar and clarity of presentation.

Units should be conform to the International System of Units (SI).

Authors can facilitate editing work by indicating in pencil, the precise meaning of certain symbols (e.g.: distinguish 0 from zero, the number 1 from the letter "l", the multiplication  $\times$  from letter X).

**Names.** Underline Latin binomials to indicate italic type.

**Figures.** Line-drawings should be clear and of high quality. Cite all figures in numerical order in the manuscript. Captions should describe the contents so that each illustration is understandable when considered apart from the text. Each illustration should be labelled with the figure number, author's name, and *Acta Agronomica*.

High-quality glossy prints of photographs should be cropped at right angles to show only essential details. Insert a scale bar where necessary to indicate magnification. Submit two sets of prints of equivalent quality.

**Tables.** The title should be self-explanatory and include enough information so that each table is intelligible without reference to the text or other tables. The title should summarize the information presented in the table without repeating the subheadings. Subheadings should be brief (abbreviations are acceptable) nonstandard ones can be explained in footnotes. Cite tables in numerical order in the manuscript. Information presented in a table should agree with that in the text.

**References.** List literature cited in alphabetic order by authors' surnames. The list should contain names and initials of all authors (et al. is not accepted here); for *journal articles* year of publication, the title of the paper, title of the journal abbreviated (do not abbreviate one word titles), volume number, first and last page. Russian titles should be transliterated and Hungarian titles translated in parentheses.

For books or chapters of books, the titles are followed by the publisher as well as place and date of publication.

Examples:

Kis, Gy., Papp, I., Bakondi-Zámori, É., Gartner-Bánfalvi, Á. (1977): A szója fungicid magcsávázásának és rhizóbiум oltásának együttes tanulmányozása (Joint study of fungicide dressing and rhizobium inoculation in soybean). *Növénytermelés*, **26**, 147-153.

Zinovev, L. S., Matalova, T. S. (1976): Protaviteli, bezopasnie dlya klubenykovykh bakterii. *Zashchita Rastenii*, **5**, 29-31.

Mather, K. and Jinks, J. L. (1971): *Biometrical genetics*. Chapman and Hall Ltd., London, U. K.



---

Periodicals of the Hungarian Academy of Sciences are obtainable  
at the following addresses:

**AUSTRALIA**

C.B.D. LIBRARY AND SUBSCRIPTION SERVICE  
Box 4886, G.P.O., *Sydney N.S.W. 2001*  
COSMOS BOOKSHOP, 145 Ackland Street  
*St. Kilda (Melbourne), Victoria 3182*

**AUSTRIA**

GLOBUS, Höchstädtplatz 3, *1206 Wien XX*

**BELGIUM**

OFFICE INTERNATIONAL DES PERIODIQUES  
Avenue Louise, 485, *1050 Bruxelles*  
E. STORY-SCIENTIA P.V.B.A.  
P. van Duyseplein 8, *9000 Gent*

**BULGARIA**

HEMUS, Bulvar Ruszki 6, *Sofia*

**CANADA**

PANNONIA BOOKS, P.O. Box 1017  
Postal Station "B", *Toronto, Ont. M5T 2T8*

**CHINA**

CNPICOR, Periodical Department, P.O. Box 50  
*Peking*

**CZECHOSLOVAKIA**

MAD'ARSKA KULTURA, Národní třída 22  
*115 66 Praha*  
PNS DOVOZ TISKU, Vinohradská 46, *Praha 2*  
PNS DOVOZ TLACE, *Bratislava 2*

**DENMARK**

EJNAR MUNKSGAARD, 35, Nørre Segade  
*1370 Copenhagen K*

**FEDERAL REPUBLIC OF GERMANY**

KUNST UND WISSEN ERICH BIBER  
Postfach 46, *7000 Stuttgart 1*

**FINLAND**

AKATEMINEN KIRJAKAUPPA, P.O. Box 128  
*00101 Helsinki 10*

**FRANCE**

DAWSON-FRANCE S.A., B.P. 40, *91121 Palaiseau*  
OFFICE INTERNATIONAL DE DOCUMENTATION ET  
LIBRAIRIE, 48 rue Gay-Lussac  
*75240 Paris, Cedex 05*

**GERMAN DEMOCRATIC REPUBLIC**

HAUS DER UNGARISCHEN KULTUR  
Karl Liebknecht-Straße 9, *DDR-102 Berlin*

**GREAT BRITAIN**

BLACKWELL'S PERIODICALS DIVISION  
Hythe Bridge Street, *Oxford OX1 2ET*  
BUMPUS, HALDANE AND MAXWELL LTD.  
Cowper Works, *Olney, Bucks MK46 4BN*  
COLLET'S HOLDINGS LTD., Denington Estate,  
*Wellingborough, Northants NN8 2QT*  
WM DAWSON AND SONS LTD., Cannon House  
*Folkstone, Kent CT19 5EE*  
H. K. LEWIS AND CO., 136 Gower Street  
*London WC1E 6BS*

**GREECE**

KOSTARAKIS BROTHERS INTERNATIONAL  
BOOKSELLERS, 2 Hippokratous Street, *Athens-143*

**HOLLAND**

FAXON EUROPE, P.O. Box 167  
*1000 AD Amsterdam*  
MARTINUS NIJHOFF B. V.

Lange Voorhout 9-11, *Den Haag*  
SWETS SUBSCRIPTION SERVICE  
P.O. Box 830, *2160 Sz Lisse*

**INDIA**

ALLIED PUBLISHING PVT. LTD.  
750 Mount Road, *Madras 600002*  
CENTRAL NEWS AGENCY PVT. LTD.  
Connaught Circus, *New Delhi 110001*  
INTERNATIONAL BOOK HOUSE PVT. LTD.  
Madame Cama Road, *Bombay 400039*

**ITALY**

D. E. A., Via Lima 28, *00198 Roma*  
INTERSCIENTIA, Via Mazzè 28, *10149 Torino*  
LIBRERIA COMMISSIONARIA SANSONI  
Via Lamarmora 45, *50121 Firenze*  
SANTO VANASIA, Via M. Macchi 58  
*20124 Milano*

**JAPAN**

KINOKUNIYA COMPANY LTD.  
Journal Department, P.O. Box 55  
*Chitose, Tokyo 156*  
MARUZEN COMPANY LTD., Book Department  
P.O. Box 5050 Tokyo International, *Tokyo 100-31*  
NAUKA LTD., Import Department  
2-30-19 Minami Ikebukuro, *Toshima-ku, Tokyo 171*

**KOREA**

CHULPANMUL, *Phenjan*

**NORWAY**

TANUM-TIDSKRIFT-SENTRALEN A.S.  
Karl Johansgata 43, *1000 Oslo*

**POLAND**

WĘGIERSKI INSTYTUT KULTURY  
Marszałkowska 80, *00-517 Warszawa*  
CKP I W, ul. Towarowa 28, *00-958 Warszawa*

**ROUMANIA**

D. E. P., *Bucuresti*  
ILEXIM, Calea Grivitei 64-66, *Bucuresti*

**SOVIET UNION**

SOYUZPECHAT — IMPORT, *Moscow*  
and the post offices in each town  
MEZHDUNARODNAYA KNIGA, *Moscow G-200*

**SPAIN**

DIAZ DE SANTOS Lagasca 95, *Madrid 6*

**SWEDEN**

ESSELTE TIDSKRIFTSCENTRALEN  
Box 62, *101 20 Stockholm*

**SWITZERLAND**

KARGER LIBRI AG, Petersgraben 31, *4011 Basel*

**USA**

EBSCO SUBSCRIPTION SERVICES  
P.O. Box 1943, *Birmingham, Alabama 35201*  
F. W. FAXON COMPANY, INC.  
15 Southwest Park, *Westwood Mass. 02090*  
MAJOR SCIENTIFIC SUBSCRIPTIONS  
1851 Diplomat, P.O. Box 819074,  
*Pallas, Tx. 75381-9074*  
READ-MORE PUBLICATIONS, INC.  
140 Cedar Street, *New York, N. Y. 10006*

**YUGOSLAVIA**

JUGOSLOVENSKA KNJIGA, Terazije 27, *Beograd*  
FORUM, Vojvode Mišića 1, *21000 Novi Sad*